Cognition in patients treated for pituitary diseases

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Introduction

Chapter 1

General Introduction

General Introduction

Pituitary diseases are characterized by abnormalities related to undersecretion or oversecretion of pituitary hormones with or without local mass effects. The cause of these diseases is often a pituitary tumor, which is usually benign and emanates from the anterior pituitary. In general treatment of pituitary adenomas consists of surgery, which may be followed by radiotherapy in cases of a significant tumor remnant or regrowth. Due to the pituitary disease itself or after treatment with surgery and/or radiotherapy, hormonal therapy may be necessary in case of hormonal disturbances. Literature suggests that these different treatment options may have an effect on cognition. However, reported results are inconsistent and mostly derived from small and heterogeneous (according to diagnoses and treatment) patient groups. The studies presented in this thesis try to unravel the effects of radiotherapy and hormonal and medical therapy on cognition in patients treated for pituitary diseases.

Pituitary function

The pituitary gland consists of an anterior (adenohypophysis) and posterior (neurohypophysis) lobe and produces hormones that affect different target organs (Figure 1). The adenohypophysis produces the following hormones:

- Adrenocorticotrophic Hormone (ACTH) stimulates the adrenal cortex to induce the synthesis and secretion of cortisol. Cortisol is essential for life and regulates or supports a wide variety of important cardiovascular, metabolic, immunologic, and homeostatic functions.
- Thyroid Stimulating Hormone (TSH) activates the thyroid gland in producing thyroid hormones (T3, T4) that are primarily responsible for the regulation of metabolism.
- Growth hormone (GH) affects the liver in secreting Insulin-like growth factor-1 (IGF-1) which has growth-stimulating effects on bone, muscle and other tissue function.
- Follicle stimulating hormone (FSH) and luteinizing hormone (LH) stimulate the gonadal glands in secreting reproductive hormones (testosterone, estrogen and progesterone) and promotes spermatogenesis in men and the growth and maturation of follicles in the ovaries in women.

The neurohypophysis produces the antidiuretic hormone (ADH) which regulates the amount of urine produced by the kidneys. A shortage of ADH can lead to disturbances in the water homeostasis.

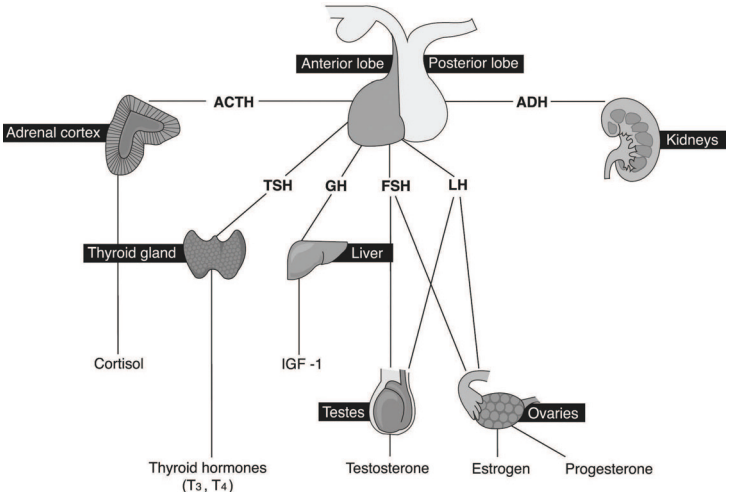

Figure 1. A selection of hormones and associated target organs produced by the pituitary.

Altogether, these hormones help us to live by regulating various metabolic and sexual functions and by allowing an adequate response to physical or emotional stress.

Pituitary diseases

Pituitary diseases can present with symptoms of hormonal excess or hormonal insufficiency or by mass effects. In the context of this thesis two conditions related to hormonal insufficiency and one condition related to hormonal excess are described.

Hormonal insufficiency

Nonfunctioning pituitary adenomas

Nonfunctioning pituitary adenomas (NFAs) are clinically inactive tumors. However, they can cause symptoms because of increasing size and pressure on the pituitary itself or structures near the pituitary such as the optic nerve or optic chiasm. As a consequence neurological symptoms like a loss of vision or double vision, or less commonly, headache can occur. When the pituitary gland itself loses one or more of its functions, the syndrome will be called hypopituitarism. Symptoms of hypopituitarism are diverse and are related to the severity of the hormonal insufficiency and the function of the specific hormones. The treatment of choice of a nonfunctioning pituitary adenoma (NFA) is surgery which may be followed by radiotherapy in cases with tumor remnant or regrowth. Additional hormonal substitution is needed for those hormones that are no longer produced. Of these, glucocorticoids (hydrocortisone and cortisone acetate) are of particular interest in this thesis, since they bind abundantly on brain receptors which are important for cognitive functioning.

Secondary adrenal insufficiency

There are many diseases that may cause dysfunctioning of the pituitary gland. Examples are mac-

roprolactinomas, cysts with pituitary localization, craniopharyngiomas, germinomas, meningeomas, Ear Nose and Throat tumors, Primitive Neuroectodermal Tumors, Sheehan's Syndrome, traumatic brain injury or congenital forms of hormone deficiency. All of the aforementioned diseases can cause secondary adrenal insufficiency.

Patients with secondary adrenal insufficiency have impaired ACTH and thus cortisol production and present with a slowly progressive weight and appetite loss, anorexia, and generalized fatigue [1]. Patients with secondary adrenal insufficiency might also suffer from a reduced quality of life and have an increased risk of mortality and morbidity compared to normal population [2,3]. Patients are treated with glucocorticoids (GC) to compensate for the loss of endogenous cortisol production. Usually this is done by oral administration of hydrocortisone or cortisone acetate aiming to mimic a normal circadian cortisol rhythm, with peak values in the early morning and low concentrations at bedtime.

Hormonal excess

Acromegaly

Acromegaly is characterized by excess GH production. Characteristic features include large fleshy lips and nose, spade-like hands, frontal skull bossing, and cranial ridges. Enlarged tongue, bones, salivary glands, thyroid, heart, liver and spleen are the effects of generalized visceromegaly [4]. Furthermore, some patients are confronted with cardiovascular complications like hypertension, occurring in 46% of patients with acromegaly [5]. Endocrine and metabolic complications consist of diabetes, hypogonadism and sleep apnea. Other comorbidities may include abnormalities of bone and joints or colon polyps [6]. The primary aim of treatment for acromegaly is to normalize GH and IGF-1 hypersecretion. Treatment of choice for most GH-producing pituitary adenomas is surgery. However, acromegaly cannot be cured in many cases because of the inability to completely remove the adenoma, necessitating other treatment modalities for disease control such as radiotherapy and antihormonal treatment.

Treatment for pituitary diseases

Surgery

Treatment for pituitary diseases commonly starts by surgical removal of the tumor, which can alleviate pressure effects and hormonal hypersecretion and may sometimes restore hormone hyposecretion. Transsphenoidal surgery is the most common way to remove the pituitary tumor. Transsphenoidal means that the tumor is approached through the sphenoidal sinus, a cavity of the sphenoid bone of the skull. In experienced hands, transsphenoidal surgery is associated with minimal morbidity and mortality [4]. Less commonly, a craniotomy is indicated when the tumor is large and is not accessible by the transsphenoidal route. A craniotomy starts by removal of a part of the skull bone and involves a more extensive surgery.

Radiotherapy

Radiotherapy may be applied when the pituitary adenoma is not fully resected after surgery, because of inaccessibility for complete resection or due to critical structures in this area. Also in cases of tumor regrowth radiotherapy is applied. In both cases (tumor remnant or regrowth),

radiotherapy improves the local control rate to 95-97% for at least a period of 10 years following radiotherapy [7-9].

In most pituitary patients, fractionated external beam radiotherapy is given with linear accelerators (4-18 MV). Radiotherapy for pituitary tumors varies according to fraction dose, total dose, irradiated volume and radiotherapy technique. Conventional external beam radiotherapy consists of two opposed lateral technique, a three-, four-, or five-field technique or a combination of these techniques. These techniques can be coplanar or non-coplanar. Coplanar means that radiation fields overlap in the same plane. Since the availability of three-dimensional radiation treatment planning systems in the 1990s, non-coplanar radiation techniques became possible. Non-coplanar means that radiation fields do not overlap in the same plane and therefore an overdose to normal tissue can be minimized.

The safety of irradiation has been questioned because of concerns related to second tumor formation, cerebrovascular disease and increased mortality [10]. Besides physical side effects, radiotherapy for pituitary tumors may also be associated with cognitive impairments [11].

Hormonal and medical therapy

Hormonal therapy can be used to treat hypo- and hyperpituitarism. Hypopituitarism reflects a decreased secretion of one or more hormones and is a main symptom in patients with a NFA. Most pituitary hormones can be replaced directly or indirectly by administering the products produced by the target organs, e.g. hydrocortisone or cortisone acetate (cortisol) for adrenal insufficiency; levothyroxine for hypothyroidism; synthetic GH in case of GH deficiency; sex hormones in case of LH or FSH deficiencies (testosterone for male hypogonadism, estradiol for female hypogonadism) and desmopressin in case of ADH deficiency.

Hyperpituitarism occurs when the pituitary adenoma produces an excess of one of the hormones. Examples are an ACTH producing adenoma causing Cushing's disease or a GH producing adenoma causing acromegaly. In case of GH excess after surgery and/or radiotherapy, acromegaly patients need GH suppressive medication, such as long-acting somatostatin analogues and/or pegvisomant [12].

The effects of treatment for pituitary diseases on cognition

Radiotherapy and cognition

Some studies found that irradiated patients with pituitary adenomas showed more impairments in the domain of executive functioning (i.e. planning, cognitive flexibility, inhibition [13]) compared with patients who received no radiotherapy [11]. However, others reported that radiotherapy was not responsible for the cognitive impairments that were documented in pituitary patients [8; 14-16].

These inconsistent study results may be attributed to heterogeneity of the patients that were included (according to pituitary pathology and treatment) and small patient samples. Furthermore, a review regarding cognitive consequences of surgery and radiotherapy for pituitary tumors, suggested that methodological issues may account for the apparent disparate cognitive data that exist in this patient group and recommended that studies should concentrate on patients with a NFA whose endocrine status is stable [17].

Other factors that might be of influence on the incidence and severity of cognitive im-

pairments in patients with pituitary disease after radiotherapy are related to the radiotherapy itself, such as fraction dose, total dose and irradiated volume. However, also other disease- and treatment related factors (such as white matter lesions) might play a role [18-20]. Detailed dosimetric radiotherapy studies in relation to objective measures of cognition are lacking in humans. These studies offer the opportunity to relate radiation exposure of prespecified brain areas to cognitive performance typically associated with these brain areas. Precise radiotherapy dose-volume reconstructions in the brain allow the comparison of radiation exposure of radiation-sensitive brain areas of different radiotherapy techniques. Many imaging studies that described the effects of radiotherapy on cognition focused on patients without fixed tumor localization [21-24]. It would be better to study brain abnormalities on imaging in relation to cognition in patients with fixed tumor localization thereby excluding potential confounding influences of the localization of both brain tumor and subsequent targeted treatment and hence cognitive functioning. Furthermore, different brain regions appear to vary with regard to the susceptibility to radiotherapy [25-27].

Acromegaly and cognition

Changes in cognitive functioning can also be expected as a consequence of GH excess or GH deficiency. The reason for this assumption is a wide distribution of binding sites for IGF-1 in the brain, in particular in the medial temporal lobe (the hippocampus) and the prefrontal cortex [28]. Indeed, studies on the effects of GH and IGF-1 performed in GH deficient patients demonstrated an association between GH and cognitive performance, where poor cognitive performance was ameliorated with GH treatment [29-31]. In acromegaly patients, cognitive functioning is also reported to be impaired [32;33]. However in previous studies comparisons were made with healthy controls which is less informative because non-specific psychological factors associated with chronic illness will undoubtedly influence the results [34]. More informative would be a comparison with patients who have a NFA. These patients share many disease characteristics with acromegaly patients but do not have the GH excess. This type of study was performed by Tiemensma *et al.*, who concluded that acromegaly patients showed no cognitive dysfunctions after long-term cure [35]. The relation of previous GH excess with impaired cognitive functioning should be further investigated by comparing acromegaly patients with patients who have a NFA.

Furthermore, in case of persistent disease after surgery with or without radiotherapy, GH suppressive medication is indicated. Until now no data are available on the cognitive effects of medical treatment in acromegaly.

Adrenal insufficiency and cognition

Cognitive impairments have been reported in healthy individuals treated with GC. There is compelling evidence of an inverted "U"-shape relation between plasma GC levels and cognitive function [36]. In particular, deficits in memory [37-44] and executive functioning [45, 46] are reported in association with higher cortisol doses in healthy volunteers. These impairments could be explained by the fact that memory and executive functioning rely on brain structures containing high concentrations of GC receptors, like the hippocampus [47] and the prefrontal cortex [48]. Indeed, chronically elevated cortisol levels are associated with reduced hippocampal volume and impairments in memory [41]. The prefrontal cortex is, however, not only associated with executive function, but

also with social cognition [49] and attention [50]. To our knowledge there are no studies examining the effects of long-term hydrocortisone replacement on cognition in patients with secondary adrenal insufficiency.

Aims of this thesis

As outlined above considerable uncertainty exists regarding the effects of treatment for pituitary diseases on cognition. Reliable information is however of particular importance for clinicians and patients to allow an informed discussion and decision on treatment strategies. Therefore we explored the effects of 1. radiotherapy on cognition in patients with a NFA, 2. current GH suppressive medical treatment on cognition in acromegaly patients and 3. hydrocortisone substitution on cognition in patients with secondary adrenal insufficiency due to various pituitary diseases.

Short outline of this thesis

Chapter 2 describes a study on cognition in a large homogeneous (according to diagnosis and type of surgery) sample of patients treated for NFAs. In this study cognitive performance of patients who received surgery and radiotherapy was compared to the cognitive performance of patients who only underwent surgery. Chapter 3 further explores the effect of radiotherapy on cognition by relating the radiation dose on radiosensitive brain areas to cognitive test performance. In addition, it was determined which radiotherapy technique was superior in limiting dose to radiotherapy sensitive brain areas. Chapter 4 assesses and compares cognition and brain abnormalities on magnetic resonance imaging (MRI) in NFA patients with and without cognitive impairments.

The second part of this thesis explores the effects of hormonal treatment for pituitary diseases on cognition. Chapter 5 describes a study on cognition in acromegaly patients with persistent disease (i.e. on GH suppressive mediation) compared to acromegaly patients who were in remission. In addition, acromegaly patients were compared to patients with a NFA, to investigate the effects of previous growth hormone excess. Chapter 6 examines cognitive functioning in patients with adrenal insufficiency. Up to now, there are no studies available on the long term effects of glucocorticoids on cognition in patients with adrenal insufficiency.

Chapter 7 presents a general discussion of the results and the conclusions of this thesis. Furthermore, methodological issues are discussed and the clinical implications deriving from this thesis are considered.

References

1 Stewart PM. The Adrenal Cortex. In: Larsen PR, Kronenberg HM, Melmed S, Polonsky KS. Williams Textbook of Endocrinology. 10th edition. Saunders. An imprint of Elsevier Science. Philadelphia, Pennsylvania, 2003, 491-552.

2 Hahner S, Loeffler M, Fassnacht M, Weismann D, Koschker AC, Quinkler M, Decker O, Arlt W, Allolio B 2007 Impaired subjective health status in 256 patients with adrenal insufficiency on standard therapy based on cross-sectional analysis. *J Clin Endocrinol Metab* 92(10): 3912-3922.

3 Bergthorsdottir R, Leonsson-Zachrisson M, Oden A, Johannsson G 2006 Premature mortality in patients with Addison's disease: A population-based study. *J Clin Endocrinol Metab* 91(12): 4849-4853.

4 Melmed S, Kleinberg DL. Anterior Pituitary. In: Larsen PR, Kronenberg HM, Melmed S, Polonsky KS. Williams Textbook of Endocrinology. 10th edition. Saunders. An imprint of Elsevier Science. Philadelphia, Pennsylvania, 2003, 177-281.

5 Vitale G, Pivonello R, Auriemma RS, Guerra E, Milone F, Savastano S, Lombardi G, Colao A 2005 Hypertension in acromegaly and in the normal population: Prevalence and determinants. *Clin Endocrinol (Oxf)* 63(4): 470-476.

6 Melmed S, Casanueva FF, Klibanski A, Bronstein MD, Chanson P, Lamberts SW, Strasburger CJ, Wass JA, Giustina A 2013 A consensus on the diagnosis and treatment of acromegaly complications. *Pituitary* 16(3): 294-302.

7 Erridge SC, Conkey DS, Stockton D, Strachan MW, Statham PF, Whittle IR, Grant R, Kerr GR, Gregor A 2009 Radiotherapy for pituitary adenomas: Long-term efficacy and toxicity. *Radiother Oncol* 93(3): 597-601.

8 McCord MW, Buatti JM, Fennell EM, Mendenhall WM, Marcus RB,Jr, Rhoton AL, Grant MB, Friedman WA 1997 Radiotherapy for pituitary adenoma: Long-term outcome and sequelae. *Int J Radiat Oncol Biol Phys* 39(2): 437-444.

9 van den Bergh AC, van den Berg G, Schoorl MA, Sluiter WJ, van der Vliet AM, Hoving EW, Szabo BG, Langendijk JA, Wolffenbuttel BH, Dullaart RP 2007 Immediate postoperative radiotherapy in residual nonfunctioning pituitary adenoma: Beneficial effect on local control without additional negative impact on pituitary function and life expectancy. *Int J Radiat Oncol Biol Phys* 67(3): 863-869.

10 Gittoes NJ 2005 Pituitary radiotherapy: Current controversies. *Trends Endocrinol Metab* 16(9):407-413.

11 Noad R, Narayanan KR, Howlett T, Lincoln NB, Page RC 2004 Evaluation of the effect of radiotherapy for pituitary tumors on cognitive function and quality of life. *Clin Oncol (R Coll Radiol)* 16(4): 233-237.

12 Melmed S 2006 Medical Progress: Acromegaly. *N Engl J Med* 355(24): 2558-2573.

13 Shallice T and Burgess P 1996 The domain of supervisory processes and temporal organization of behaviour. Philosophical Transactions of the Royal Society of London. *Series B, Biological Sciences,* 351, 1405-1411.

14 Guinan EM, Lowy C, Stanhope N, Lewis PD, Kopelman MD 1998 Cognitive effects of pituitary tumors and their treatments: Two case studies and an investigation of 90 patients. *J Neurol Neurosurg Psychiatry* 65(6):870-876.

15 Peace KA, Orme SM, Padayatty SJ, Godfrey HP, Belchetz PE 1998 Cognitive dysfunction in patients with pituitary tumor who have been treated with transfrontal or transsphenoidal surgery or medication. *Clin Endocrinol (Oxf)* 49(3): 391-396.

16 Peace KA, Orme SM, Thompson AR, Padayatty S, Ellis AW, Belchetz PE 1997 Cognitive dysfunction in patients treated for pituitary tumors. *J Clin Exp Neuropsychol* 19(1): 1-6.

17 Tooze A, Gittoes NJ, Jones CA, Toogood AA 2009 Neurocognitive consequences of surgery and radiotherapy for tumors of the pituitary. *Clin Endocrinol (Oxf)* 70(4): 503-511.

18 Lawrence YR, Li XA, el Naqa I, Hahn CA, Marks LB, Merchant TE, Dicker AP 2010 Radiation dose-volume effects in the brain. *Int J Radiat Oncol Biol Phys* 76(3 Suppl): S20-7.

19 Armstrong CL, Gyato K, Awadalla AW, Lustig R, Tochner ZA 2004 A critical review of the clinical effects of therapeutic irradiation damage to the brain: The roots of controversy. *Neuropsychol Rev* 14(1): 65-86.

20 Armstrong CL, Hunter JV, Ledakis GE, Cohen B, Tallent EM, Goldstein BH, Tochner Z, Lustig R, Judy KD, Pruitt A, Mollman JE, Stanczak EM, Jo MY, Than TL, Phillips P 2002 Late cognitive and radiographic changes related to radiotherapy: Initial prospective findings. *Neurology* 59(1): 40-48.

21 Surma-aho O, Niemela M, Vilkki J, Kouri M, Brander A, Salonen O, Paetau A, Kallio M, Pyykkonen J, Jaaskelainen J 2001 Adverse long-term effects of brain radiotherapy in adult low-grade glioma patients. *Neurology* 56(10) :1285-1290.

22 Postma TJ, Klein M, Verstappen CC, Bromberg JE, Swennen M, Langendijk JA, Taphoorn MJ, Scheltens P, Slotman BJ, van der Ploeg HM, Aaronson NK, Heimans JJ 2002 Radiotherapyinduced cerebral abnormalities in patients with low-grade glioma. *Neurology* 59(1): 121-123.

23 Douw L, Klein M, Fagel SS, van den Heuvel J, Taphoorn MJ, Aaronson NK, Postma TJ, Vandertop WP, Mooij JJ, Boerman RH, Beute GN, Sluimer JD, Slotman BJ, Reijneveld JC, Heimans JJ 2009 Cognitive and radiological effects of radiotherapy in patients with low-grade glioma: Long-term follow-up. *Lancet Neurol* 8(9): 810-818.

24 Meyers CA, Hess KR 2003 Multifaceted end points in brain tumor clinical trials: Cognitive deterioration precedes MRI progression. *Neuro Oncol* 5(2): 89-95.

25 Gondi V, Tome WA, Mehta MP 2010 Why avoid the hippocampus? A comprehensive review. *Radiother Oncol* 97(3): 370-376.

26 Marsh JC, Gielda BT, Herskovic AM, Abrams RA 2010 Cognitive sparing during the administration of whole brain radiotherapy and prophylactic cranial irradiation: Current concepts and approaches. *J Oncol* 198208.

27 Redmond KJ, Achanta P, Grossman SA, Armour M, Reyes J, Kleinberg L, Tryggestad E, Quinones-Hinojosa A, Ford EC 2011 A radiotherapy technique to limit dose to neural progenitor cell niches without compromising tumor coverage. *J Neurooncol* 104(2): 579-587.

28 van Dam PS, Aleman A 2004 Insulin-like growth factor-I, cognition and brain aging. *Eur J Pharmacol* 490(1-3): 87-95.

29 Sathiavageeswaran M, Burman P, Lawrence D, Harris AG, Falleti MG, Maruff P, Wass J 2007 Effects of GH on cognitive function in elderly patients with adult-onset GH deficiency: A placebo-controlled 12-month study. *Eur J Endocrinol* 156(4): 439-447.

30 Oertel H, Schneider HJ, Stalla GK, Holsboer F, Zihl J 2004 The effect of growth hormone substitution on cognitive performance in adult patients with hypopituitarism. *Psychoneuroendocrinology* 29(7): 839-850.

31 Falleti MG, Maruff P, Burman P, Harris A 2006 The effects of growth hormone (GH) deficiency and GH replacement on cognitive performance in adults: A meta-analysis of the current literature. *Psychoneuroendocrinology* 31(6): 681-691.

32 Leon-Carrion J, Martin-Rodriguez JF, Madrazo-Atutxa A, Soto-Moreno A, Venegas-Moreno E, Torres-Vela E, Benito-Lopez P, Galvez MA, Tinahones FJ, Leal-Cerro A 2010 Evidence of cognitive and neurophysiological impairment in patients with untreated naive acromegaly. *J Clin Endocrinol Metab* 95(9): 4367-4379.

33 Sievers C, Samann PG, Pfister H, Dimopoulou C, Czisch M, Roemmler J, Schopohl J, Stalla GK, Zihl J 2012 Cognitive function in acromegaly: Description and brain volumetric correlates. *Pituitary* 15(3): 350-357.

34 van den Berg E, Kloppenborg RP, Kessels RP, Kappelle LJ, Biessels GJ 2009 Type 2 diabetes mellitus, hypertension, dyslipidemia and obesity: A systematic comparison of their impact on cognition. *Biochim Biophys Acta* 1792(5): 470-481.

35 Tiemensma J, Biermasz NR, van der Mast RC, Wassenaar MJ, Middelkoop HA, Pereira AM, Romijn JA 2010 Increased psychopathology and maladaptive personality traits, but normal cognitive functioning, in patients after long-term cure of acromegaly. *J Clin Endocrinol Metab* 95(12): E392-402.

36 Gallagher P, Reid KS, Ferrier IN 2009 Neuropsychological functioning in health and mood disorder: Modulation by glucocorticoids and their receptors. *Psychoneuroendocrinology* 34 Suppl 1: S196-207.

37 de Quervain DJ, Roozendaal B, Nitsch RM, McGaugh JL, Hock C 2000 Acute cortisone administration impairs retrieval of long-term declarative memory in humans. *Nat Neurosci* 3(4): 313-314.

38 Kirschbaum C, Wolf OT, May M, Wippich W, Hellhammer DH 1996 Stress- and treatment induced elevations of cortisol levels associated with impaired declarative memory in healthy adults. *Life Sci* 58(17): 1475-1483

39 Lupien SJ, Gaudreau S, Tchiteya BM, Maheu F, Sharma S, Nair NP, Hauger RL, McEwen BS, Meaney MJ 1997 Stress-induced declarative memory impairment in healthy elderly subjects: Relationship to cortisol reactivity. *J Clin Endocrinol Metab* 82(7): 2070-2075.

40 Lupien SJ, Wilkinson CW, Briere S, Ng Ying Kin NM, Meaney MJ, Nair NP 2002 Acute modulation of aged human memory by pharmacological manipulation of glucocorticoids. *J Clin Endocrinol Metab* 87(8): 3798-3807.

41 Lupien SJ, Fiocco A, Wan N, Maheu F, Lord C, Schramek T, Tu MT 2005 Stress hormones and human memory function across the lifespan. *Psychoneuroendocrinology* 30(3): 225-242.

42 Newcomer JW, Craft S, Hershey T, Askins K, Bardgett ME 1994 Glucocorticoid-induced impairment in declarative memory performance in adult humans. *J Neurosci* 14(4): 2047-2053.

43 Newcomer JW, Selke G, Melson AK, Hershey T, Craft S, Richards K, Alderson AL 1999 Decreased memory performance in healthy humans induced by stress-level cortisol treatment. *Arch Gen Psychiatry* 56(6): 527-533.

44 Alhaj HA, Massey AE, McAllister-Williams RH 2008 Effects of cortisol on the laterality of the neural correlates of episodic memory. *J Psychiatr Res* 42(12): 971-981.

45 Lupien SJ, Gillin CJ, Hauger RL 1999 Working memory is more sensitive than declarative memory to the acute effects of corticosteroids: A dose-response study in humans. *Behav Neurosci* 113(3): 420-430.

46 McCormick CM, Lewis E, Somley B, Kahan TA 2007 Individual differences in cortisol levels and performance on a test of executive function in men and women. *Physiol Behav* 91(1): 87-94.

47 Joels M 2008 Functional actions of corticosteroids in the hippocampus. *Eur J Pharmacol* 583(2-3): 312-321.

48 Sanchez MM, Young LJ, Plotsky PM, Insel TR 2000 Distribution of corticosteroid receptors in the rhesus brain: Relative absence of glucocorticoid receptors in the hippocampal formation. *J Neurosci* 20(12): 4657-4668

49 Van Overwalle F 2009 Social cognition and the brain: A meta-analysis. *Hum Brain Mapp* 30(3): 829-858.

50 Li C, Chen K, Han H, Chui D, Wu J 2012 An FMRI study of the neural systems involved in visually cued auditory top-down spatial and temporal attention. *PLoS One* 7(11): e49948.

Radiotherapy and the effects on cognition

Chapter 2

Cognitive functioning in patients treated for nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy

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Abstract

Context and objective Cognitive deterioration is reported in patients with a nonfunctioning pituitary macroadenoma (NFA) and after pituitary radiotherapy. However, reported results are inconsistent and are potentially confounded by different underlying pituitary disorders. The aim of this study was to examine cognitive functions in patients previously treated for NFA with or without radiotherapy.

Design Verbal memory was assessed with the Dutch equivalent to the Rey Auditory Verbal Learning Test (15 Words Test, 15 WT). Executive functioning was examined using the Ruff Figural Fluency Test (RFFT). We compared our patient cohort with large reference populations representative of the Dutch population.

Patients Eighty-four patients (62 ± 10 years) who underwent transsphenoidal surgery 8.6 ± 6.3 years earlier participated. Patients who underwent radiotherapy (n = 39) were compared to those who received surgery alone (n = 45). All patients were on stable hormonal replacement therapy.

Results The total patient group scored significantly below the reference sample on all 15 WT *Z-scores* (95%CI): short-term memory, -0.3 (-0.5 to -0.1); total memory, -0.8 (-1.1 to -0.5); learning score, -0.3 (-0.5 to -0.1); delayed memory, -0.8 (-1.1 to -0.5), all $P < 0.01$. The total patient group scored significantly below the reference sample on RFFT *Z-scores* (95%CI): unique designs, -0.7 (-0.9 to -0.5) and perseverative errors, -0.5 (-0.8 to -0.2), both $P < 0.001$. Patients who underwent radiotherapy showed no significant differences on cognition when compared to those who received surgery alone.

Conclusion Patients with NFA score significantly worse on cognition compared to reference populations. Radiotherapy does not appear to have a major influence on cognition.

Introduction

Nonfunctioning pituitary macroadenomas (NFAs) are clinically inactive tumors. The treatment of choice of NFAs is transsphenoidal surgery followed by external beam radiotherapy (RT) in cases with tumor remnant or regrowth, thereby improving the local control rate to 95-97% for at least a period of 10 years following RT [1-3].

Although effective, the safety of irradiation has been questioned because of concerns related to second tumor formation, cerebrovascular disease and increased mortality [4]. Besides physical side effects, RT of pituitary tumors has been associated with cognitive impairments. Noad *et al.* compared aspects of memory, language processing, visual-spatial abilities and executive functions (i.e. planning, cognitive flexibility, inhibition [5]) between patients who received either surgery for pituitary tumors or a combination of surgery and RT. A comparison between patients' performances with reference data indicated that both outcome groups show cognitive impairments on visual-spatial abilities and immediate memory. However, patients in the radiotherapy group scored significantly worse on executive functioning compared to patients without radiotherapy [6]. Other studies found cognitive impairments in pituitary patients where RT did not seem to be responsible [2;7-9]. Furthermore, we previously found that irradiated patients with NFA did not report more cognitive problems than patients with NFA following surgery alone [10]. Although self-rated questionnaires are very helpful in clinical medicine to rapidly assess possible disturbances in cognitive functioning, they are unrelated to objective cognitive performance [11-13].

The inconsistent previous study results may be attributed to the heterogeneity of the studied patients (according to pituitary pathology) and small patient samples. Also, in a recent review regarding neurocognitive consequences of surgery and radiotherapy for pituitary tumors, it was suggested that methodological issues may account for the apparent disparate neurocognitive data that exist in this patient group and recommended that studies should concentrate on patients with a NFA whose endocrine status is stable [14].

Reliable information on the possible side effects of RT is of particular importance for clinicians and patients to allow an informed discussion and decision on treatment strategies. Therefore, the aim of this study was to assess objective measures of memory and executive functioning in a large homogeneous (according to diagnosis and type of surgery) sample of patients with NFA. The second aim was to compare patients who had surgery and radiotherapy with those who only underwent surgery. Memory and executive functioning were examined because they have been shown to be particularly sensitive to the effects of treatment (surgery and or RT) in patients with pituitary adenomas [2;6-9].

Patients and methods

Patients

In this cross-sectional study, patients were recruited for participation at the endocrine outpatient clinic of the University Medical Center Groningen (UMCG), a tertiary referral center for pituitary surgery in the Netherlands. Inclusion criteria were age ≥ 18 years, treatment for a NFA and regular follow-up in our endocrine outpatient clinic (i.e. at least once a year). The diagnosis of NFA was based on two criteria: the presence of a pituitary macroadenoma (>1 cm) on imaging and the

absence of overproduction of any of the pituitary hormones. Pituitary deficiencies were defined according to generally accepted guidelines. In short, insufficiency of the HPA axis was based on 8 a.m. cortisol concentration < 230 mmol/l [15] and, in case of doubt, an insulin tolerance test (ITT). Thyroid hormone deficiency was based on a low serum fT4 concentration (<11.0 pmol/l). Growth hormone deficiency was based on a low IGF-1 *Z-score* less than -2 SD and/or an insufficient peak GH concentration (< 10 mU/l) in response to insulin-induced hypoglycaemia or a peak growth hormone < 18 mU/l during an arginine-GHRH test (< 18mU/l). Insufficiency of the pituitary – gonadal axis was defined in men as a testosterone concentration below 10 nmol/l, in premenopausal women (aged < 50 years) as loss of menses and in postmenopausal women (aged > 50 years) as LH and FSH concentrations below 15 mU/l. Diabetes insipidus was defined as the incapacity to properly concentrate urine (increased urine volume with low urine osmolality) in the face of a high plasma osmolality (and sodium). Biochemical control of adequacy of hormonal substitution treatment was judged by the physicians that were responsible for the care of the participating patients using fT4, IGF-1 and testosterone levels where necessary. All patients included in the present analysis underwent transsphenoidal surgery as primary treatment, in some cases followed by a second surgical procedure if a large remnant, amenable to surgery, persisted. We report data of patients who underwent transsphenoidal surgery, because this is a standard initial treatment in most cases. Furthermore, patients who underwent a craniotomy more often had larger tumors, necessitating more aggressive treatment affecting postoperative comorbidity and potentially cognitive performance. To assure that acute post-treatment effects had resolved, patients were only included at least 6 months after surgery or radiotherapy, and their hormone replacement schedules had to be stable during these preceding months. Patients were not eligible for participation if they had a neurological or psychiatric condition, if they had impairments of vision or hearing or a restriction in hand function which was expected to interfere with test performance. In addition, patients were not eligible when they were recently diagnosed with a chronic disease or a depressive disorder (as indicated by treating physician letters) and in case of pregnancy or an addictive disorder. Patients were asked by telephone to participate prior to their regular visit at our endocrine clinic, and the study procedures were performed subsequent to the regular outpatient visit. Of a total 173 patients with NFA visiting our endocrine clinic, 84 consecutive patients who received transsphenoidal surgery as primary treatment were tested between September 2008 and December 2009. The baseline characteristics of the entire cohort (n = 173) did not differ from the presented study population, confirming the representativeness of our study population (data not shown). Approval was given by the medical ethics review committee of the UMCG.

Radiotherapy

Fractionated external beam RT was given with linear accelerators (4-18 megavolts) between 1984 and 2008 (n = 39). Conventional external beam RT was administered using a two-opposed lateral technique, a three-, four-, or five-field technique or a combination of these techniques. The daily radiation fraction size varied from 1.8 to 2.0 Gray (Gy); the total radiation dose ranged from 45 to 55.8 Gy with a mean of 45.8 Gy. Average treatment time was 36 days (range 32 - 61 days). In the time period from 1984 to 1990, the radiation dose to the tumor was prescribed at the tumor encompassing isodose (n = 8). From 1991 onward, the radiation dose was prescribed at a central

point in the tumor according to the recommendations of the Internal Commission on Radiation Units and Measurements at the UMCG (n = 31).

Cognitive tests

Aspects of verbal memory were assessed with the 15 Words Test (15 WT) which is a Dutch equivalent of the Rey Auditory Verbal Learning Test [16]. In this test, 15 words were presented five times. After each trial, patients were asked to name immediately the words they remember. This allows the calculation of three different scores describing *immediate memory:*

1. The short-term memory score is based on the number of words patients were able to name after the first presentation of the word list.
2. The total memory score represents the total number of words patients remembered over the five trials.
3. The learning score describes the difference between the number of words remembered in the third trial in comparison with the first trial.

Besides immediate memory, *delayed memory* was measured.

4. The delayed recall memory score is based on the number of words patients could recall after a period of about 30 minutes.

Executive functioning was assessed using the Ruff Figural Fluency Test (RFFT) [17]. In this test, patients were presented with sheets of paper on which 35 squares were printed, each with a fixed pattern of five dots. The test consisted of five parts which differed with regard to the designs. While the configurations of dots are the same in the first three parts of the test, two types of distractions are added in two of these parts. In the last two parts, the configurations of the dots are different and without distractions. The participant was asked to produce as many different designs as possible by connecting two or more dots in each square with straight lines. The time for each part was restricted to 1 minute so that the total test time was 5 minutes. Responses were scored with regard to the total number of *unique designs* generated over the five parts. The *perseverative errors score* represents the total number of repetitions of the same design drawn. The inter-rater variability (two independent raters) was determined by Pearson's *r* and was 0.99 for both total unique designs and persevarative errors. The *error ratio* is the total number of *perseverative errors* divided by the total number of unique designs. As previous research demonstrated that performances on fluency tasks may be influenced by the use of strategies [18], clustering and switching strategies were also analyzed. This is referred to as qualitative analysis. *Clusters* were defined as being of a length of at least two consecutive related designs. According to Tucha *et al.*, clusters in structured figural fluency tasks such as the RFFT can be analyzed based on addition, subtraction and rotation strategies [18]. Clusters that contained errors were counted when at least two valid designs were included. However, errors were not calculated with regard to cluster size. *Mean cluster size* was determined by dividing the number of unique designs within clusters by the *number of clusters* generated. Because there is the possibility that clusters overlap, designs could belong to several clusters simultaneously. Beside the mean cluster size, the length of *the largest cluster* was also registered. Finally, the *number of switches* was analyzed. A switch was defined as the change between two clusters that follow immediately after each other. The inter-rater variabilities (two independent raters) were determined by Pearson's *r* and ranged from 0.85 to 0.98. All of the aforementioned scores were calculated over the whole test period of 5 minutes.

Questionnaires and protocol

A common questionnaire on demographic and health-related data was used with special attention for educational level, social status, full-time/part-time employment, social security benefit, comorbidity, use of medicine, cardiovascular risk factors, traumatic brain injury and dementia. Education level was determined by using a Dutch education system, comparable to the ISCED (International Standard Classification of Education [19]). This scale ranges from 1 (elementary school not finished) to 7 (university level). The Hospital Anxiety and Depression Scale (HADS) consists of 14 items and measures anxiety and depression [20]. Each item is scored as a number, with a maximum score for each subscale (anxiety or depression) of 21. Higher scores indicate more severe anxiety or depression.

In fixed order, the test protocol was as follows: (1) the 15 WT: direct recall, (2) the RFFT, (3) a common questionnaire to assess baseline information, (4) physical examination: length, weight, blood pressure, waist circumference, hip circumference and compliance to the test situation, (5) the HADS and finally (6) the 15 WT: delayed recall. The assessment took approximately forty minutes and was performed directly after or just before visit to the outpatient clinic. All testing and scoring of tests were performed by trained personnel.

Reference data: healthy control subjects

The performances of patients were compared to Dutch controls. Normative data for the HADS were derived from Spinhoven *et al.* [20]. In their study, psychometric properties of the HADS were assessed in six different groups of Dutch subjects (n = 6165): (1) a random sample of younger adults (18-65 years) (n = 199); (2) a random sample of elderly subjects of 57 to 65 years of age (n = 1901); (3) a random sample of elderly subjects of 66 years and older (n=3293); (4) a sample of consecutive general practice patients (n=112); (5) a sample of consecutive general medical outpatients with unexplained somatic symptoms (n=169); and (6) a sample of consecutive psychiatric out-patients (n=491). In all six groups, an authorized Dutch translation of the HADS was used. General population mean and standard deviations scores were used from 18 to 65 years and >65 years to calculate *Z-scores*. Reference data for the 15 WT were derived from control subjects of the Maastricht Aging Study [16]. In this study, 1780 healthy participants between 24 and 81 years were evaluated on a Dutch adaptation of the Rey Verbal Learning Test (RVLT), the 15 WT. Regressions models given by the authors were used to determine the exact *Z-scores*. The final test scores were controlled for age, sex, and education.

Reference data for the RFFT were derived from a sample (n = 10,289) of the LifeLines Cohort Study [21]. Reference groups are stratified by a matrix of 8 education levels and 13 age levels (half decades from 20 to 85 years). Each cluster consisted on average of 120 subjects. RFFT forms were analyzed by a computerized pattern recognition program. There was a good internal consistency (n = 373) between computerized rating and human rating for unique designs (Cronbach's α = 0.99) and perseverative errors (Cronbach's α = 0.97). Using a Bland Altman analysis, a near perfect level of agreement was found between these two rating methods with intraclass correlation for unique designs: 0.99 and perseverative errors: 0.96. Using the mean and standard deviation scores for each reference group, we standardized our patient scores by converting into *Z-scores*.

Statistical analyses

The analyses were all carried out using the SPSS package for Windows version 16.0.2. Statistical analysis between patient groups was performed using t-tests. Categorical variables were analyzed by using chi-square tests. The following data are presented in tables: (1) Raw scores (Mean ± SD), (2) the number and percentage of patients scoring below the median (i.e. ≤ 50th percentile on the basis of reference data) and (3) the number and percentage of patients scoring impaired. According to Lezak, cognitive impairment on a test was defined as a performance equivalent to or below the 10th percentile of the reference samples (equivalent to a *Z-score* of ≤ -1.3) [22]. For graphical representation, study population mean *Z-scores* with 95% confidence intervals is shown. Because some of the variables were not normally distributed, nonparametric tests were used to confirm the findings of parametric tests. The nonparametric analysis supported the results of the parametric tests; therefore, only the results of the parametric tests are reported. The two-tailed alpha level of < 0.05 was considered as statistically significant. In case of statistical differences between groups for cognitive test results, a Bonferroni correction was done to correct for multiple testing.

Results

Study population

Eighty-four patients (55 men and 29 women, aged 62 ± 10 years, range 36 to 81 years) participated in the present study. Thirty-nine patients received transsphenoidal surgery and postoperative RT (RT+ group), whereas 45 patients did not receive RT (RT- group). Patients' characteristics are given in Table 1. Patients who underwent RT were significantly younger at the time of surgery and their duration of follow-up from first surgery was longer. In addition, the irradiated patients used more hormonal substitution, and this reached significance for the use of adrenal and thyroid hormones. Average time between first surgery and radiotherapy was approximately 17 months. Feelings of anxiety and depression were comparable between the RT+ and RT- group and not indicative of clinical anxiety and depression. Social status, full-time/part-time employment, social security benefit and comorbidity were all comparable between both outcome groups at the time of assessment (data not shown).

Cognitive tests

The total patient group scored significantly lower on the 15 WT compared to the reference sample, *Z-scores* (95% confidence interval, *P* level): short-term memory, -0.3 (-0.5 to -0.1, $P = 0.009$); total memory, -0.8 (-1.1 to -0.5, $P < 0.001$); learning score, -0.3 (-0.5 to -0.1, $P = 0.006$); delayed memory, -0.8 (-1.1 to -0.5, $P < 0.001$). However, there were no significant differences between the RT+ and RT- group (Table 2, Figure 1).

The total patient group scored significantly lower on the RFFT for the measures of unique designs and perseverative errors compared to the reference sample, *Z-scores* (95% confidence interval, *P* level): unique designs, -0.7 (-0.9 to -0.5, $P < 0.001$); perseverative errors, -0.5 (-0.8 to -0.2, $P < 0.001$).

There was a trend in the error ratio, -0.3 (-0.6 to 0.01, $P = 0.058$). Comparisons between the RT+ and RT- group showed no significant differences on executive functioning for both qualitative and quantitative analyses (Table 2, Figure 1).

***Table 1**. Clinical characteristics of patients treated for nonfunctioning pituitary macroadenoma*

	Total	RT+	RT-	
N	84	39	45	*P*-value*
Basic characteristics				
Age (y)	62 (10)	60 (11)	63 (9)	0.234
Sex (males/females)	55/29	25/14	30/15	0.805
Educational level (1/2/3/4/5/6/7)	1/8/1/18/37/16/3	1/3/1/11/18/3/2	0/5/0/7/19/13/1	0.142
Surgery				
Age at surgery (y)	52.9 (12.4)	48.5 (12.8)	56.8 (10.8)	0.002
Average time since surgery (y)	8.6 (6.3)	11.7 (7.0)	5.9 (4.0)	<0.001
< 1 – 5 years (number, (%))	31 (37)	10 (26)	21 (47)	<0.001
5 – 10 years	24 (29)	7 (18)	17 (38)	
> 10 years	29 (35)	22 (56)	7 (16)	
Patients with 2nd surgery (%)	11	15	7	0.198
Radiotherapy				
Age at radiotherapy (y)		50 (13)		
Time between primary surgery and RT (y)		1.4 (1.5)		
Hormonal substitution				
No. of hormone replacements (0/1/2/3/4/5)	11/17/13/24/19/0	1/8/4/17/9/0	10/9/9/7/10/0	0.010
Glucocorticoid (%)	64	77	53	0.023
Thyroid hormone (%)	66	82	51	0.003
Growth hormone (%)	31	39	24	0.116
Sex hormone (%)	55	54	56	0.875
Desmopressin (%)	12	13	11	0.809

RT+: group of patients who underwent surgery and radiotherapy
RT-: group of patients who underwent surgery alone
* RT+ versus RT-

Table 2. Cognitive performance of patients treated for nonfunctioning pituitary macroadenoma

	Total	RT+	RT-	
N	84	39	45	*P*-value*
15 Words Test				
Short-term memory				
Mean (SD)	4.6 (1.9)	4.4 (1.9)	4.7 (1.9)	0.467
≤ 50th perc. (%)	67	69	64	0.643
≤10th perc. (%)	20	26	16	0.251
Total memory				
Mean (SD)	37.6 (11.3)	36.2 (11.6)	38.8 (10.9)	0.294
≤ 50th perc. (%)	76	85	69	0.091
≤10th perc. (%)	37	46	29	0.102
Learning score				
Mean (SD)	3.2 (2.0)	3.0 (2.0)	3.3 (2.0)	0.419
≤ 50th perc. (%)	63	72	56	0.124
≤10th perc. (%)	19	23	16	0.381
Delayed memory				
Mean (SD)	7.1 (3.4)	7.3 (3.5)	6.9 (3.4)	0.600
≤ 50th perc. (%)	76	74	78	0.714
≤10th perc. (%)	38	39	38	0.949
Ruff Figural Fluency Test				
Unique designs				
Mean (SD)	58.5 (25.1)	55.7 (29.0)	60.9 (21.2)	0.352
≤ 50th perc. (%)	74	74	73	0.915
≤10th perc. (%)	37	44	31	0.237
Perseverative errors				
Mean (SD)	9.9 (10.9)	7.8 (8.2)	11.7 (12.6)	0.098
≤ 50th perc. (%)	80	82	78	0.627
≤10th perc. (%)	18	21	16	0.554
Error ratio				
Mean (SD)	0.2 (0.2)	0.2 (0.2)	0.2 (0.2)	0.411
≤ 50th perc. (%)	76	85	69	0.091
≤10th perc. (%)	13	15	11	0.563
Clustering & Switching				
Mean cluster size (SD)	2.7 (0.7)	2.7 (0.7)	2.7 (0.8)	0.385
Mean no. clusters (SD)	12.2 (6.2)	11.3 (6.7)	13.0 (5.7)	0.175
Mean no. switches (SD)	3.9 (3.5)	3.6 (3.8)	4.1 (3.3)	0.427
Largest Cluster (SD)	4.6 (2.0)	4.6 (2.1)	4.7 (2.1)	0.352

RT+: group of patients who underwent surgery and radiotherapy
RT-: group of patients who underwent surgery alone
* RT+ versus RT-.

Figure 1. *Z-scores of verbal memory (15 Words Test) and executive functioning (RFFT) in the RT+ and RT- group.*

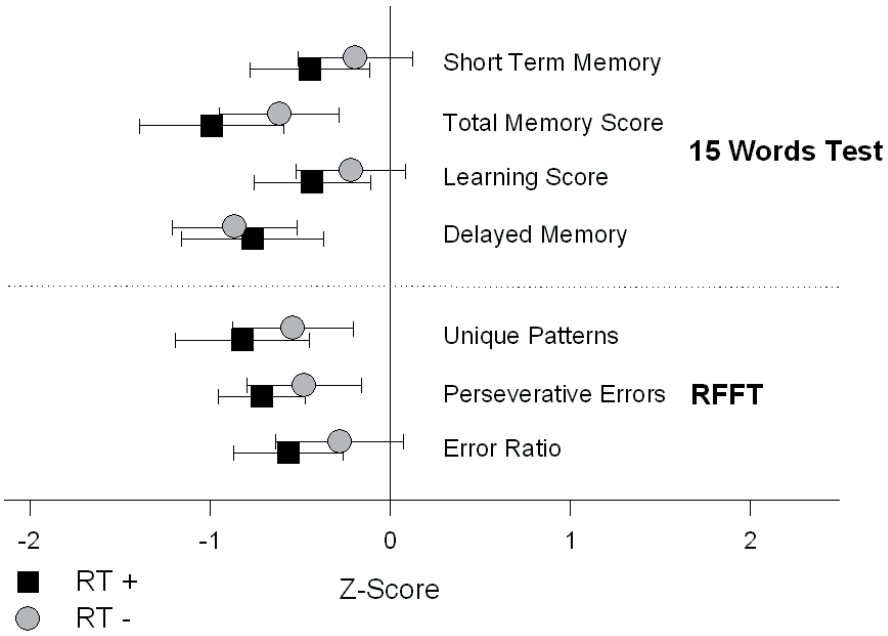

Figure 1
Data are mean *Z-scores* and 95% confidence intervals
RT+: group of patients who underwent surgery and radiotherapy
RT-: group of patients who underwent surgery alone
RFFT: Ruff Figural Fluency Test

Discussion

This study shows that patients treated for NFAs scored significantly worse than reference populations on both verbal memory and executive functioning. However, no significant differences were found between patients who received radiotherapy compared to patients who underwent surgery alone.

Our finding is in agreement with most studies mentioned before, even though most of them used heterogeneous patient groups with different tumor types [2;6-9;23]. Recently, Tiemensma *et al.* described cognitive functioning in Cushing and NFA patients [24]. Although the main scope of this article was on irreversible effects of previous hypercortisolism, comparisons of patients with NFA and matched controls were made. The NFA patient group scored significantly below the matched control group on one memory subtest and on two tests of executive functioning. However, the authors do not describe whether they corrected for multiple testing. Furthermore, no significant

differences were found on global cognitive functioning. For this latter assessment, the Mini Mental State Examination was used, which is known to be rather insensitive due to its ceiling effect [25]. The results of this study are also comparable to those of Guinan *et al.* who performed a large and detailed study [7]. They found anterograde memory deficits (i.e. difficulties in learning new information) in all patient groups (acromegaly, Cushing's disease, NFAs and prolactinoma) when compared to control subjects. There were no effects of different treatment modalities (type of surgery, radiotherapy or use of medication) on memory scores.

In a study by McCord *et al.*, distinct differences were reported compared to our results [2]. They found significantly more severe memory problems in the RT+ group compared to the RT- group. However, the patient group was heterogeneous according to tumor etiology and treatment. Another limitation of their study might be that patients had to rate their memory problems themselves by filling out a questionnaire. Self-rated questionnaires must be interpreted with caution. Numerous studies that focused on the subjective experience of cognitive functioning have consistently found that self-reports are unrelated to objective performance in distinct patient groups [11-13]. Noad *et al*. found that irradiated patients performed significantly worse than patients treated with surgery alone on an executive functioning test [6]. The discrepancy between the study of Noad *et al.* and the results of this study might be explained by the fact that different tests for executive functioning were used. Executive functioning is a broad term, encompassing e.g. inhibition, cognitive flexibility and planning. While Noad *et al.* used a test that relied mainly on the capability of inhibition, the RFFT relies mainly on planning capacities. As different cognitive functions rely on different brain networks, which could be affected more or less because of radiotherapy, future studies on the effects of radiotherapy should aim to correlate radiation field and dose to cognitive performance. This type of studies has not been performed in patients who received pituitary radiotherapy. These studies will provide insight into potential radiation-induced brain damage and actual cognitive performance. Although we found no significant differences between the RT+ and the RT- group, the RT+ patients almost always scored below the RT- group. There is a possibility that brain-induced damage by radiotherapy is very subtle, and therefore larger patient groups are required. A retrospective power analyses revealed that at least 392 patients in each group would have been required to detect significant differences in both tests used with 80% power, illustrating that the effects of RT are at most very small and unlikely to be of clinical relevance.

Although we did not find significant differences between the RT+ and RT- group, the total group scored significantly worse compared to reference data. This can be a consequence of transsphenoidal surgery. Peace *et al*. found that nearly one-third of patients who underwent this type of surgery (without radiotherapy) had memory and executive test scores below the 10th percentile compared to < 5% of the controls. In addition, they found that many nonsurgical patients suffer from mild cognitive impairments [9]. Thus, it is likely that the primary disease or hormonal abnormalities secondary to the tumor and/or its treatment could be responsible. Over the last decades, evidence has accumulated, indicating that glucocorticoïds play an important role in the regulation of memory [26]. Moreover, research in congenital hypothyroid patients or hypothyroidism in adulthood has shown the importance of thyroid hormone for cognition [27;28]. However, we found no effects on cognitive performance of glucocorticoïd or thyroid substitution therapy

in multivariate analysis within our study population (data not shown). Our hormonal substitution therapy appeared to be adequate (i.e. hormonal *Z-scores* in the normal range) despite its intrinsic imperfections. Alternatively, cognitive impairments could be a result of nonspecific psychological factors associated with having a chronic illness. A recent systematic review found that type 2 diabetes, hypertension and, to a lesser extent, obesity and dyslipidemia are also associated with mild-to-moderate decrements in cognitive functioning in nondemented persons. The profile of cognitive decrements was nonspecific, with most consistent results found in the domains of memory, processing speed and cognitive flexibility [29].

Age at surgery is another potential risk factor that might have influenced the results. Younger age at surgery could be a favorable recovery factor after brain surgery. This could be because of a more neuroplastic brain at younger age and special (vascular) vulnerability in the elderly patients [30]. In our study, the RT+ group was younger than the RT- group at pituitary surgery and as a result had a longer average time since surgery. However, both age at surgery and time since surgery or radiotherapy were not associated with cognitive functioning (data not shown).

In conclusion, in patients treated for nonfunctioning pituitary adenomas memory and executive functioning are impaired. This could not be attributed to any treatment modality, including pituitary radiotherapy.

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References

1 van den Bergh AC, van den Berg G, Schoorl MA, Sluiter WJ, van der Vliet AM, Hoving EW, Szabó BG, Langendijk JA, Wolffenbuttel BH, Dullaart RB. 2007 Immediate postoperative radiotherapy in residual nonfunctioning pituitary adenoma: beneficial effect on local control without additional negative impact on pituitary function and life expectancy. I*nt J Radiat Oncol Biol Phys* 67(3): 863-9.

2 McCord MW, Buatti JM, Fennell EM, Mendenhall WM, Marcus RB, Jr., Rhoton AL, Grant MB, Friedman WA. 1997 Radiotherapy for pituitary adenoma: long-term outcome and sequelae. *Int J Radiat Oncol Biol Phys*, 39(2): 437-44.

3 Erridge SC, Conkey DS, Stockton D, Strachan MW, Statham PF, Whittle IR, Grant R, Kerr GR, Gregor A. 2009 Radiotherapy for pituitary adenomas: long-term efficacy and toxicity. *Radiother Oncol* 93(3): 597-601.

4 Gittoes NJ. 2005 Pituitary radiotherapy: current controversies. *Trends Endocrinol Metab* 16(9): 407-13.

5 Shallice T, Burgess P. 1996 he domain of supervisory processes and temporal organization of behaviour. *Philos Trans R Soc Lond B Biol Sci* ;351(1346): 1405-11.

6 Noad R, Narayanan KR, Howlett T, Lincoln NB, Page RC. 2004 Evaluation of the effect of radiotherapy for pituitary tumors on cognitive function and quality of life. *Clin Oncol (R Coll Radiol)* 16(4): 233-7.

7 Guinan EM, Lowy C, Stanhope N, Lewis PD, Kopelman MD. 1998 Cognitive effects of pituitary tumors and their treatments: two case studies and an investigation of 90 patients. *J Neurol Neurosurg Psychiatry* 65(6): 870-6.

8 Peace KA, Orme SM, Thompson AR, Padayatty S, Ellis AW, Belchetz PE. 1997 Cognitive dysfunction in patients treated for pituitary tumors. *J Clin Exp Neuropsychol* 19(1): 1-6.

9 Peace KA, Orme SM, Padayatty SJ, Godfrey HP, Belchetz PE. 1998 Cognitive dysfunction in patients with pituitary tumor who have been treated with transfrontal or transsphenoidal surgery or medication. *Clin Endocrinol (Oxf)* 49(3): 391-6.

10 van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, Langendijk JA, Wolffenbuttel BH. 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated for nonfunctioning pituitary adenoma. *Int J Radiat Oncol Biol Phys* 68(4): 986-91.

11 Maor Y, Olmer L, Mozes B. 2001 The relation between objective and subjective impairment in cognitive function among multiple sclerosis patients-the role of depression. *Mult Scler* 7(2): 131-5.

12 Middleton LS, Denney DR, Lynch SG, Parmenter B. 2006 The relationship between perceived and objective cognitive functioning in multiple sclerosis. *Arch Clin Neuropsychol* 21(5): 487-94.

13 Sawrie SM, Martin RC, Kuzniecky R, Faught E, Morawetz R, Jamil F, Viikinsalo M, Gilliam F. 1999 Subjective versus objective memory change after temporal lobe epilepsy surgery. *Neurology* 53(7): 1511-7.

14 Tooze A, Gittoes NJ, Jones CA, Toogood AA. 2009 Neurocognitive consequences of surgery and radiotherapy for tumors of the pituitary. *Clin Endocrinol (Oxf)* 70(4): 503-11.

15 Dullaart RP, Pasterkamp SH, Beentjes JA, Sluiter WJ. 1999 Evaluation of adrenal function in patients with hypothalamic and pituitary disorders: comparison of serum cortisol, urinary free cortisol and the human-corticotrophin releasing hormone test with the insulin tolerance test. *Clin Endocrinol (Oxf)* 50(4): 465-71.

16 van der Elst W, Van Boxtel MP, Van Breukelen GJ, Jolles J. 2005 Rey's verbal learning test: normative data for 1855 healthy participants aged 24-81 years and the influence of age, sex, education, and mode of presentation. *J Int Neuropsychol Soc* 11(3): 290-302.

17 Ruff RM, Light RH, Evans RW 1987 The Ruff Figural Fluency Test: A normative study with adults. *Developmental Neuropsychology*, 3, 37-51.

18 Tucha O, Mecklinger L, Laufkotter R, Kaunzinger I, Paul GM, Klein HE, Lange KW. 2005 Clustering and switching on verbal and figural fluency functions in adults with attention deficit hyperactivity disorder. *Cogn Neuropsychiatry* 10(3): 231-48.

19 United Nations Educational, Scientific and Cultural Organization. International Standard Classification of Education ISCED. 2006.

20 Spinhoven P, Ormel J, Sloekers PP, Kempen GI, Speckens AE, Van Hemert AM 1997 A validation study of the Hospital Anxiety and Depression Scale (HADS) in different groups of Dutch subjects. *Psychol Med* 27(2): 363-70.

21 Stolk RP, Rosmalen JG, Postma DS, de Boer RA, Navis G, Slaets JP, Jormel J, Wolffenbuttel BH. 2008 Universal risk factors for multifactorial diseases: LifeLines: a three-generation population-based study. *Eur J Epidemiol* 23(1): 67-74.

22 Lezak, MD, Havieson DB, Loring DW 2004 Neurospsychological Assessment (4th Ed.).

23 Grattan-Smith PJ, Morris JG, Shores EA, Batchelor J, Sparks RS. 1992 Neuropsychological abnormalities in patients with pituitary tumors. *Acta Neurol Scand* 86(6): 626-31.

24 Tiemensma J, Kokshoorn NE, Biermasz NR, Keijser BJ, Wassenaar MJ, Middelkoop HA, Pereira AM, Romijn JA. 2010 Subtle cognitive impairments in patients with long-term cure of Cushing's disease. *J Clin Endocrinol Metab* 95(6): 2699-714.

25 Spreen O, Strauss E. 1998 A Compendium of Neuropsychological Tests: Administration, Norms, and Commentary.

26 de Quervain DJ, Aerni A, Schelling G, Roozendaal B. 2009 Glucocorticoids and the regulation of memory in health and disease. *Front Neuroendocrinol* 30(3): 358-70.

27 Morreale de Escobar G, Obregon MJ, Escobar del Rey F. 2004 Role of thyroid hormone during early brain development. *Eur J Endocrinol* 151 Suppl 3: U25-U37.

28 Osterweil D, Syndulko K, Cohen SN, Pettler-Jennings PD, Hershman JM, Cummings JL, Tourtellotte WW, Solomon DH. 1992 Cognitive function in non-demented older adults with hypothyroidism. *J Am Geriatr Soc* 40(4): 325-35.

29 van den Berg E, Kloppenborg RP, Kessels RP, Kappelle LJ, Biessels GJ. 2009 Type 2 diabetes mellitus, hypertension, dyslipidemia and obesity: A systematic comparison of their impact on cognition. *Biochim Biophys Acta* 1792(5): 470-81.

30 Mussig K, Leyhe T, Besemer B, Saur R, Häring HU, Gallwitz B, Klinberg S. 2009 Younger age is a good predictor of better executive function after surgery for pituitary adenoma in adults. *J Int Neuropsychol Soc* 15(5): 803-6.

Chapter 3

Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex

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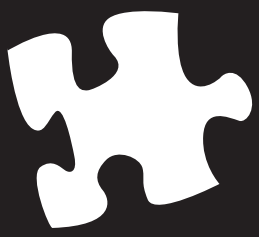

Abstract

Objective The hippocampus and prefrontal cortex (PFC) are important for memory and executive functioning and are known to be sensitive to radiotherapy (RT). Radiation dosimetry relates radiation exposure to specific brain areas. The effects of various pituitary RT techniques were studied by relating detailed dosimetry of the hippocampus and PFC to cognitive performance.

Methods In this cross-sectional design, 75 nonfunctioning pituitary macroadenoma (NFA) patients (61 ± 10 years) participated and were divided into irradiated (RT+, n = 30) and non-irradiated (RT-, n = 45) groups. The RT+ group (who all received 25 fractions of 1.8 Gray; total dose: 45 Gray) consisted of three RT technique groups; three-field technique, n = 10; four-field technique, n=15 and five-field technique, n=5. Memory and executive functioning were assessed by standardized neuropsychological tests. A reconstruction of the dose distributions for the three RT techniques was made. The RT doses on 30, 50 and 70% of the volume of the left and right hippocampus and PFC were calculated.

Results Cognitive test performance was not different between the four groups, despite differences in radiation doses applied to the hippocampi and PFC. Age at RT, time since RT, and the use of thyroid hormone varied significantly between the groups; however, they were not related to cognitive performance.

Conclusion This study showed that there were no significant differences on cognitive performance between the three-, four- and five-field RT groups and the non-irradiated patient group. A dose-response relationship could not be established, even with a radiation dose that was higher on most of the volume of the hippocampus and PFC in case of a four-field RT technique compared with the three- and five-field RT techniques.

Introduction

Patients with a nonfunctioning pituitary macroadenoma (NFA) may receive postoperative radiotherapy (RT) for local control [1;2]. However, the safety of RT to the brain has been questioned because of concerns related to second tumor induction, cerebrovascular disease, and increased mortality [3]. In addition, accelerated cognitive decline may develop over many years following RT. The incidence and severity of this complication is dependent on radiation fraction dose, total dose, and volume, but can also be influenced by other disease- and treatment-related factors [4]. Susceptibility of different brain regions is also reported to vary [5-7]. In addition, time since RT is considered to be an important factor because deterioration in cognitive functions might appear only after a few years [8]. Furthermore, both younger and older patients carry a greater risk of cognitive impairment from RT [9].

Patients who received RT for primary brain tumors performed worse on tests for executive functioning [10]. Other groups reported poor memory performance in irradiated low-grade glioma patients [11]. However, controversy remains because studies on the effects of RT on cognition are usually difficult to interpret, giving differences in tumor localization, which are likely to affect various cognitive domains. In addition, Armstrong *et al.* described that radiation effects appear to be severe only in a minority of patients. Further, risk of cognitive impairment was found to be related to direct and indirect effects of cancer type, concurrent clinical factors, and premorbid risk factors [9].

In pituitary adenoma patients, Noad *et al.* [12] found that patients who received postoperative RT performed worse on executive functioning when compared with patients who underwent surgery alone. However, we [13] and others [14-17] did not find an effect of RT on the cognitive performance of patients treated for pituitary disease.

Although older literature supports a role for RT-induced cognitive decline, it remains to be established whether modern pituitary RT techniques result in poorer cognitive performance. However, detailed dosimetric RT studies in relation to objective measures of cognition are lacking in humans. These studies offer the opportunity to relate radiation exposure of prespecified brain areas to cognitive performance. Precise RT dose–volume reconstructions in the brain allow the comparison of radiation exposure of radiation-sensitive brain areas of different RT techniques. In this context, the temporal lobe/hippocampus and the prefrontal cortex (PFC) appear to be especially relevant. The temporal lobe is crucial for acquisition of new information as well as its storage and retrieval. The hippocampus within the temporal lobe is important for declarative memory, i.e. the conscious recollection of facts and events [18]. Specifically, the left and right hippocampus appear to have different memory functions [19;20]. The hippocampal granule cell layer, which undergoes neural regenesis, seems to be more sensitive to RT than glial or neural cells in the brain, as shown in animal models [21]. The PFC is of great importance for executive functioning (i.e. planning, cognitive flexibility and inhibition [22]) [23], which seems to decrease after RT [10;12].

Previously, we found no major influence of pituitary RT on cognition in patients with NFA [24]. However, smaller effects could not be excluded. Therefore, we decided to refine the strategy to relate the radiation dose to radiosensitive brain areas (i.e. the hippocampus and PFC) to cognitive test performance typically associated with these brain areas. In addition, we studied which RT technique was superior at limiting dose to the hippocampus and PFC.

Methods and Materials

Patients

In this cross-sectional study, patients were recruited for participation at the endocrine outpatient clinic of the University Medical Center Groningen (UMCG), a tertiary referral center for pituitary surgery in the Netherlands. Inclusion criteria were age ≥ 18 years, treatment for NFA, and regular follow-up in our endocrine outpatient clinic (i.e. at least once a year). The diagnosis of NFA was based on two criteria: the presence of a pituitary macroadenoma (>1 cm) on magnetic resonance imaging (MRI) and the absence of overproduction of any of the pituitary hormones. Pituitary deficiencies were defined according to generally accepted guidelines. Biochemical control of adequacy of the hormonal substitution treatment was judged by the physicians responsible for the care of participating patients, with the use of free thyroxine, insulin-like growth factor 1, and testosterone measurements where necessary. All patients included in the present analysis underwent transsphenoidal surgery (TSS) as a primary treatment, in some cases followed by a second surgical procedure if a large remnant, accessible only by surgery, persisted. We only report data of patients who underwent TSS because this is a standard initial treatment in most cases. Patients who underwent a craniotomy more often had larger tumors, necessitating more aggressive treatment, which affect postoperative comorbidity and potentially also cognitive performance. To assure that acute post-treatment effects had resolved, patients were included only at least 6 months after surgery or RT. Furthermore, their hormone replacement schedules had to be stable during these preceding months. Patients were not eligible for participation if they had a (prior) neurological or psychiatric condition, if they had impairments of vision or hearing, or a restriction in hand function, which was expected to interfere with test performance. In addition, patients were not eligible when they were recently diagnosed with a chronic disease or a depressive disorder, indicated by letters of the attending physicians, and in case of pregnancy or an addictive disorder. Prior to their regular visit at our endocrine clinic, patients were approached by telephone to participate. Of a total of 173 NFA patients visiting our endocrine clinics, 75 consecutive patients who received TSS as primary treatment with or without a three-, four- or five- field RT techniques were tested between September 2008 and December 2009. The baseline characteristics of the entire cohort (n = 173) did not differ from the presented study population, confirming the representativeness of our study population (data not shown). Approval was given by the medical ethics review committee of the UMCG.

Table 1. Clinical characteristics of patients treated for nonfunctioning pituitary macroadenoma (NFA) with different radiotherapy (RT) techniques (three, four, and five fields) and without RT (RT-). Data are median and interquartile range, absolute numbers or percentage.

	Three fields	Four fields	Five fields	RT-	*P*-value*
N	10	15	5	45	
Basic characteristics					
Age (y)	61 (58-71)	59 (53-70)	55 (46-61)	63 (57-70)	0.236
Sex (males/females)	9/1	10/5	3/2	30/15	0.491
Educational level (1/2/3/4/5/6/7)	1/1/0/1/5/1/1	0/2/0/5/6/1/1	0/0/0/2/2/1/0	0/5/0/7/19/13/1	0.407
Surgery					
Age at surgery (y)	48 (45-56)	53 (47-63)	42 (27-48)	57 (51-65)	0.008[a]
Average time since surgery (y)	13 (10-15)	5 (3-8)	14 (12-19)	6 (2-8)	<0.001[b]
< 1 – 5 years (number, (%))	0	8 (53)	0	21 (47)	<0.001
5 – 10 years	1 (10)	6 (40)	0	17 (38)	
> 10 years	9 (90)	1 (7)	5 (100)	7 (16)	
Patients with 2nd surgery (%)	2 (20)	1 (7)	1 (20)	3 (7)	0.478
Radiotherapy (RT)					
Age at RT (y)	48 (45-58)	55 (50-64)	43 (29-50)	NA	0.040[c]
Average time since RT (y)	13 (10-14)	3 (2-4)	12 (11-17)	NA	<0.001[d]
Hormonal substitution No. of hormone replacements (0/1/2/3/4/5)	0/1/0/6/3/0	1/3/2/5/4/0	0/2/1/1/1/0	10/9/9/7/10/0	0.218
Glucocorticoid (%)	80	80	40	53	0.119
Thyroid hormone (%)	100	67	80	51	0.026
Growth hormone (%)	50	33	40	24	0.418
Sex hormone (%)	70	60	40	56	0.712
Desmopressin (%)	10	13	20	11	0.940

*: *P*-value between the four groups by Kruskal-Wallis or Chi-square tests. NA: not applicable.
Duncan's post hoc test:
a: Significant differences between the five-field group on the hand and the four-field and RT- groups on the other hand.
b: Significant differences between the three-field and five-field groups on the one hand and the four-field and RT- groups on the other hand.
c: Significant differences between the four-field and five-field groups.
d: Significant differences between the three-field and five-field groups on the one hand and the four-field group on the other hand.

Radiotherapy

Fractionated external beam RT was given with linear accelerators (4-18 megavolts) between 1987 and 2008 (n = 30). In this time period, pituitary RT was performed using a three-, four-, or five-fields techniques (Table 1). The three-field technique replaced the two lateral opposed field technique because the older two-field technique irradiated a large volume of normal brain with an equivalent or even higher dose of what was applied to the tumor, with reports of brain necrosis as a result of that. The three-field technique consisted of two lateral fields and one vertex field. Since the availability of three-dimensional (3D) radiation treatment planning systems in the 1990s of the previous century, non-coplanar radiation techniques became possible. With non-coplanar techniques, RT fields do not overlap in the same plane and therefore overdose to normal tissue can be minimized. The three- and five-field irradiation techniques used in this study were co-planar, but the four-field technique was non-coplanar. This four-field technique was planned to spare the temporal lobes.

For all the three multiple field RT techniques, similar dose prescriptions were used: 1.8 Gray (Gy) with a total dose of 45 Gy given in 25 fractions. The median overall radiation treatment time was 35 days (range 31- 37 days). From 1987 to 1990, the RT dose of the tumor/pituitary adenoma was prescribed at the tumor encompassing isodose (n = 2). From 1991 onwards, the RT dose was prescribed at a central point in the tumor/pituitary adenoma according to the recommendations of the Internal Commission on Radiation Units and Measurements (n = 28).

The dose distribution effects of the three different RT techniques were reconstructed using a planning computed tomography (CT) and MRI scan of the head of a 64-year-old patient that served as a model for the whole patient group. The gross tumor volume (i.e. pituitary adenoma remnant) and brain areas at risk (i.e. the left and right hippocampus and PFC) were delineated on MRI with a slice thickness of 1 mm. A planning target volume was generated by adding a 3D margin of 10 mm around the gross tumor volume. The normal tissues at risk were delineated with the help of an experienced neuroradiologist (L C Meiners) according to the established neuroanatomical bounderies [25-27]. To allow a direct comparison, the 3D conformal RT treatment plans were generated using the Pinnacle treatment planning system (version 8.0h). The 3D dose distribution of the left and right hippocampus and PFC were based on a 3D reconstruction of the CT scan. For a description of the RT exposure of the left and right hippocampus and the PFC, the RT dose received at 30, 50 and 70% of these brain area volumes are given. These percentages were chosen because at 30, 50 and 70%, the largest differences between the three different RT techniques in radiation dose were seen. These percentage points give a better idea of the dose distributions compared with mimimum, maximum, mean, and SDS.

Cognitive tests

Aspects of verbal memory were assessed with the 15 Words Test (15 WT) which is a Dutch equivalent of the Rey Auditory Verbal Learning Test [28]. In this test, 15 words were presented five times. After each trial, patients were asked to name immediately the words they remembered. This allowed the calculation of three different scores describing immediate memory:

1. The short-term memory score is based on the number of words patients were able to name after the first presentation of the word list.

2. The total memory score represents the total number of words patients remembered over the five trials.
3. The learning score describes the difference between the number of words remembered in the third trial in comparison with the first trial.
Besides immediate memory, delayed memory was measured.
4. The delayed memory score is based on the number of words patients could recall after a period of about 30 minutes.

Executive functioning was assessed using the Ruff Figural Fluency Test (RFFT) [29]. In this test, patients were presented with sheets of paper on which 35 squares were printed, each with a fixed pattern of five dots. The test consisted of five parts, which differed with regard to the designs. While the configurations of dots are the same in the first three parts of the test, two types of distractions are added in two of these parts. In the last two parts, the configurations of the dots are different and without distractions. The participant was asked to produce as many different designs as possible by connecting two or more dots in each square with straight lines. The time for each part was restricted to 1 minute so that the total test time was 5 minutes. Responses were scored with regard to the total number of unique designs generated over the five parts. The perseverative errors score represents the total number of repetitions of the same design drawn. The interrater variability (two independent raters) was determined by Pearson's *r* and was 0.99 for both total unique designs and perseverative errors. The error ratio is calculated by the total number of perseverative errors divided by the total number of unique designs.

Questionnaires and protocol

A common questionnaire on demographic and health-related data was used with special attention for educational level, social status, full-time/part-time employment, social security benefit, comorbidity, use of medicine, cardiovascular risk factors, traumatic brain injury, and dementia. Education level was determined by using a Dutch education system, comparable to the International Standard Classification of Education (ISCED) [30]. This scale ranges from 1 (elementary school not finished) to 7 (university level). The Hospital Anxiety and Depression Scale (HADS) consists of 14 items and measures anxiety and depression [31]. Each item is scored as a number, with a maximum score for each subscale (anxiety or depression) of 21. Higher scores indicate more severe anxiety or depression.

In fixed order, the test protocol was as follows: (1) the 15 WT: direct recall, (2) the RFFT, (3) a common questionnaire to assess baseline information, (4) physical examination: length, weight, blood pressure, waist circumference, hip circumference and compliance to the test situation, (5) the HADS and finally (6) the 15 WT: delayed recall. The assessment took approximately forty minutes and was performed directly after or just before patients' visit to the outpatient clinic. All testing and scoring of tests were performed by trained personnel.

Reference data: healthy control subjects

The performances of patients were compared with Dutch controls. Normative data for the HADS were derived from Spinhoven *et al.* [31]. In their study, psychometric properties of the HADS were assessed in six different groups of Dutch subjects (n = 6165): (1) a random sample of younger

adults (18-65 years) (n = 199); (2) a random sample of elderly subjects of 57 to 65 years of age (n = 1901); (3) a random sample of elderly subjects of 66 years and older (n = 3293); (4) a sample of consecutive general practice patients (n = 112); (5) a sample of consecutive general medical outpatients with unexplained somatic symptoms (n = 169); and (6) a sample of consecutive psychiatric outpatients (n = 491). In all six groups, an authorized Dutch translation of the HADS was used. General population mean and SDS were used from 18 to 65 years and > 65 years to calculate *Z-scores*.

Reference data for the 15 WT were derived from control subjects of the Maastricht Aging Study. In this cohort, 1780 healthy participants between 24 and 81 years were evaluated on a Dutch adaptation of the Rey Verbal Learning Test, the 15 WT [28]. Regression models given by the authors were used to determine accurate *Z-scores*. The final test scores were controlled for age, sex, and education. Reference data for the RFFT were derived from a sample (n = 10,289) of the LifeLines Cohort Study [32]. Reference groups were stratified by a matrix of eight education levels and 13 age levels (half decades from 20 to 85 years). Each cluster consisted on average of 120 subjects. RFFT forms were analyzed by a computerized pattern recognition program. There was a good internal consistency (n = 373) between computerized rating and human rating for unique designs (Cronbach's $\alpha = 0.99$) and perseverative errors (Cronbach's $\alpha = 0.97$). Using a Bland-Altman analysis, a near perfect level of agreement was found between these two rating methods with intraclass correlation for unique designs: 0.99 and perseverative errors: 0.96. Using the mean and SDS for each reference group, we standardized our patient scores by converting it into *Z-scores*.

Statistical analyses

The analyses were all carried out using the PASW (SPSS, Inc., Armonk, NY, USA) statistics 18 package. Demographic data are presented as median and interquartile range (IQR), frequencies or percentages. We compared data of four groups of patients: 1) patients who received TSS and a three-field RT technique, 2) patients who received TSS and a four-field RT technique, 3) patients who received TSS and a five-field RT technique and 4) patients who only received TSS. Categorical variables were analyzed by using Chi-square tests. The non-parametric Kruskal-Wallis test was used for all continuous variables that failed to meet the normality assumption. The two-tailed alpha level of < 0.05 was considered statistically significant. In case of statistical differences between the groups on demographic or cognitive data, Duncan's method was used as a *post-hoc* test [33].

Results

Study population

Seventy-five NFA patients (52 men and 23 women, age 61 ± 10 years) participated in this study. Thirty patients received TSS and postoperative pituitary RT (RT+ group), whereas 45 patients did not receive RT (RT- group). The RT+ group consisted of three RT technique groups; three-field technique, n = 10; four-field technique, n = 15; five-field technique, n = 5. Patients' characteristics are given in Table 1. No differences in age at the time of study, sex, or educational level were found between the four groups. In our cohort, the older three- and five-field RT techniques were

applied earlier in time, and from 2001, these techniques were replaced by a four-field technique. Consequently, significant differences between groups were found for average time since RT. Furthermore, patients in the four-field group were on average older at time of RT compared with those in the five-field group (Table 1). Hormonal substitution for pituitary deficiencies was similar between groups with the exception of thyroid hormone that was given less frequently in patients without RT. Feelings of anxiety and depression were comparable between the four groups and not indicative of clinical anxiety and depression (data not shown). Social status, full-time/part-time employment, social security benefit, and comorbidity were all comparable between the four groups at the time of assessment (data not shown).

Radiotherapy

The RT dose distributions on transversal, coronal and sagittal CT scan images are shown for different RT techniques (Figure 1). Estimated RT dosimetric data (derived from Dose-Volume Histograms, plots not shown) revealed increased radiation dose exposure in the four-field RT technique: doses received at 50 and 70% of the hippocampus and PFC were upto seven fold higher compared with the three- and five-field RT techniques (Table 2). Furthermore, the patient model used in this study had a tumor that had a deviation to the right. Therefore, slightly higher doses were found on the right hippocampus compared with the left hippocampus.

Cognitive tests

Cognitive functioning *Z-scores* are given in Table 2 for memory performance and executive functioning. No significant differences were found on the 15 WT and the RFFT between the four groups; no RT dose-volume effect on cognition was found. Longer time since RT (three- and five-field techniques) or older age at RT (four-field technique) did not result in poorer cognitive test results.

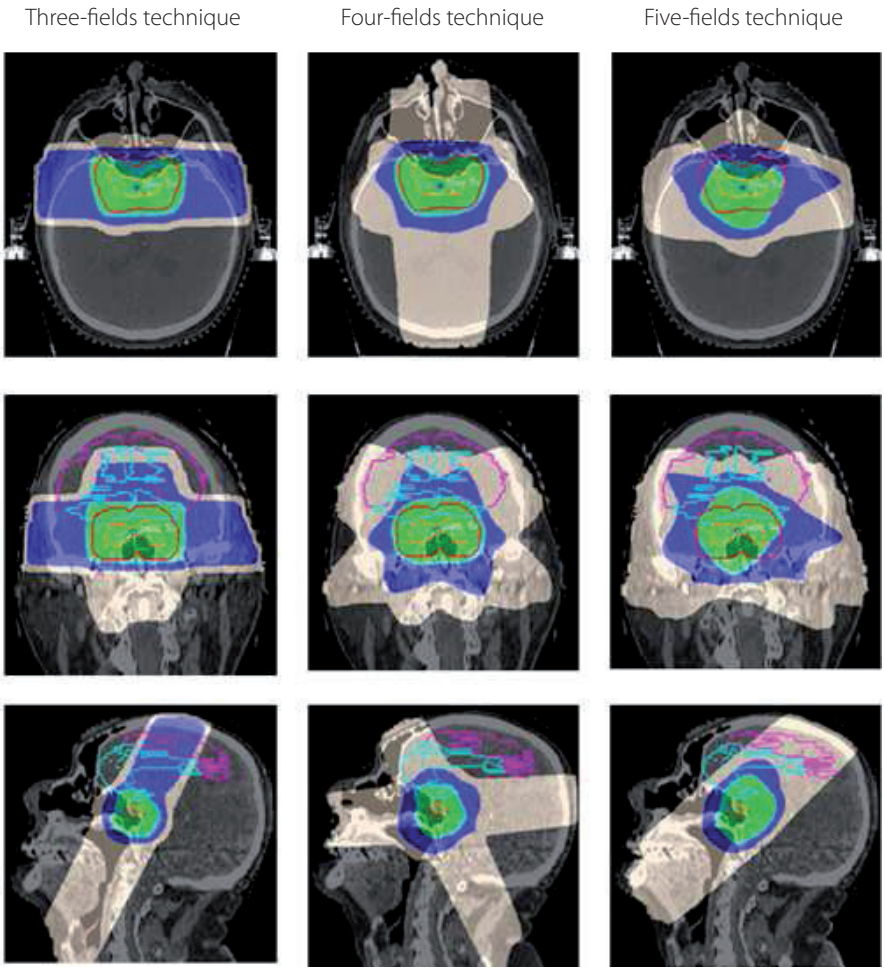

Figure 1 *The RT dose distributions on transversal, coronal, and sagittal CT scan images are shown for different RT techniques. The color areas shown on the CT scan images represent different RT isodose areas: red = 49.5-51.8 Gy; orange = 48.2 Gy; green = 42.8 – 45 Gy; light blue = 40 Gy; dark blue = 20 – 30 Gy; and white = 5-10 Gy. The color lines shown on the CT scan images represent different delineated structures: red line, planning target volume; yellow line, gross tumor volume/ pituitary adenoma remnant; light blue line, prefrontal cortex; and pink line, frontal cortex.*

Table 2 *Estimated dosimetric data memory performance and executive functioning of patients treated for nonfunctioning pituitary macroadenoma (NFA) with different radiotherapy (RT) techniques (three, four and five fields) and without RT.*

	With different RT techniques			Without RT	*P*-value*
	Three fields	Four fields	Five fields		
N	10	15	5	45	
Estimated dosimetric data (RT dose received at 30/50/70% of the volume of:)					
Left hippocampus (Gy)	23.0/3.2/2.0	21.8/14.9/13.5	19.6/4.1/2.9		
Right hippocampus (Gy)	28.5/4.4/2.0	29.8/15.2/14.2	19.0/4.6/2.8		
Prefrontal cortex (Gy)	23.0/19.0/3.6	26.5/18.2/17.3	25.6/16.2/4.9		
Memory performance (15 Words Test; mean (SD))					
Short-term memory	-0.13 (1.42)	-0.65 (0.90)	-0.39 (1.23)	-0.20 (1.09)	0.536
Total memory	-0.46 (1.66)	-1.33 (1.07)	-0.92 (0.72)	-0.62 (1.15)	0.337
Learning score	-0.23 (1.17)	-0.70 (0.93)	-0.16 (0.90)	-0.22 (1.04)	0.280
Delayed memory	0.00 (1.39)	-0.96 (1.21)	-1.26 (0.67)	-0.86 (1.19)	0.249
Executive functioning (Ruff Figural Fluency test; mean (S.D.))					
Unique designs	-0.52 (1.08)	-1.19 (1.08)	-0.43 (1.39)	-0.56 (1.13)	0.150
Perseverative errors	-0.87 (0.73)	-0.60 (0.89)	-0.61 (0.67)	-0.33 (1.46)	0.618
Error ratio	-0.80 (0.48)	-0.31 (1.40)	-0.59 (0.71)	-0.09 (1.78)	0.732

**P*-values by Kruskal-Wallis ANOVA. Cognitive performance data are given in *Z*-scores.

Discussion

This study showed that there were no significant differences between the three-, four- and five field RT groups and the non-irradiated patient group. Therefore, a dose-response relationship could not be established, even with a radiation dose that was higher on most of the volume of the hippocampus and PFC in case of a four-field RT technique compared with the three- and five-field techniques.

To our knowledge, this is the first study that related detailed dosimetric data of pituitary RT to cognitive performance. Our results confirm our previous data [13;24] and that of others [14-17] that modern dose regimens of pituitary RT does not appear to have a major influence on cognition, but also extend these by showing an absence of a relationship between radiation dose and cognitive performance. For our study, we used a homogeneous patient group of patients with NFA, thereby excluding confounding results by excess hormone or other treatment-related factors [34]. Further, our study lacks the inherent weakness of studies in patients with primary brain tumors or gliomas. Because of the fixed position of the pituitary tumor that is not affecting cortical or subcortical areas of the brain, cognitive test results were not influenced by localization of both brain tumor and subsequent targeted focal RT.

This study showed a larger cumulative radiation dose on most of the volume of the left and right hippocampus and PFC with a four-field technique. Although this radiation technique was developed to circumvent excess radiation exposure to the brain - especially both temporal lobes - by avoiding radiation fields overlap, our reconstruction showed that this technique is not preferred with regard to RT exposure to the hippocampus and PFC. Despite this excess radiation, no poorer cognitive performance was observed in this patient group. Arguably, patients with four-field RT had a shorter follow-up time (3 years) when compared with the three- and five-field techniques (13 and 12 years respectively). For this reason, cognitive dysfunction may not have developed. However, at slightly higher radiation doses, some have reported a decrease in executive functioning already after 6 months in patients with primary brain tumors [10]. It is evident that long-term follow-up is necessary for this patient group [4].

In contrast to our study, Noad *et al*. found differences between irradiated and non-irradiated pituitary patients with regard to cognitive functioning. They tested 71 patients with pituitary tumors treated with surgery with or without RT (25 fractions of 1.8 Gy) on quality of life and several cognitive functions (memory, attention and executive functioning) [12]. In addition to an impairment in cognitive function regardless of treatment type, they reported a significantly worse performance on executive function (measured with the Stroop test) in the RT+ group compared with patients receiving only surgery. As noted by the authors, their finding may be explained by chance. Jalali *et al.* [35] also found that RT applied to the left temporal lobe in patients with tumors of low malignant potential (craniopharyngioma, cerebellar astrocytoma, optic pathway glioma and cerebral low-grade glioma) were predictive of cognitive decline. In addition, Douw *et al.* [36] report a decline in attentional functioning in patients with low-grade gliomas who received RT. Among the many differences between these studies and ours, the most explanatory are the higher radiation doses used (> 54 Gy in 30 fractions), the different tumor pathology, and the young age of the patient group (median 13 years). Comparison with the above mentioned

studies [10,35,36] suggests that there may be a kind of threshold for RT to injure the brain and cause cognitive impairment. It is likely that only above this threshold a dose-response relationship can be found. The age difference between the patients of Jalali's study and ours is probably also essential, in that a developing brain is more likely to be affected by RT. The absence of a RT dose–cognitive response relationship in our study suggests that all applied RT techniques seem to operate within safe RT dose boundaries.

In accordance with our finding that pituitary RT was not associated with reduced cognition, others also found no differences between patients treated for pituitary tumors with or without RT [14-17]. In a recent review by Loeffler & Shih [37], it was stated that the overall rate of treatment-related adverse effects (secondary tumors, visual complications, neurological symptoms, strokes, general, and mental health) is low and that only hypopituitarism is to be expected following RT in pituitary adenoma patients. However, there are indications that this risk does not exceed the risk on hypopituitarism in patients who only received surgery [2]. Unfortunately, cognition received little attention in this recently published paper by Loeffler. However, Lawrence *et al.* [4] recently reviewed the published data regarding RT-induced brain injury and found very limited evidence that brain RT in 2 Gy fractions causes irreversible cognitive decline in adults with primary and metastatic brain tumors.

Some study limitations are to be made. In this study, only a small number of patients were included in some subgroups yielding low statistical power and potentially excluding detection of differences that were not large. This study showed that there were no significant differences on cognitive test performance between the three-, four- and five field RT groups and the non-irradiated patient group. Therefore, it seems plausible that RT might at most have some very subtle impact on cognition. However, some reservations are justified. Ideally, to detect such a small size effect, a prestudy analytical design with regard to group sizes should be made. Literature provides no data on estimated RT-induced cognitive decline in humans with fractionated RT limited to a total dose of 45 Gy given in 25 fractions. In addition, the disease prevalence is low (especially considering that not all received RT), creating little room for extensive prestudy analytical design. Instead, we approached all patients that were eligible within September 2008 and December 2009, which resulted in a current population size, which is relatively small, and therefore has statistical limitations.

Further, it should also be taken into account that it is plausible that not everyone shares the same vulnerability to damage of RT. For instance, Armstrong *et al.* [9] reviewed that the apolipoprotein E *(ApoE)* genotype could prove to be a premorbid risk factor for greater RT-induced damage, possibly related to its risk for the development of neurofibrillary tangles and plaque neuritis [38]. In our group, we found no differences in *ApoE* genotype between the four groups with normal allele frequencies of apoE4. Finally, to precisely define the volume of the hippocampus and PFC, we used imaging of a 64-year-old patient. The exact volume of the hippocampus and PFC differs inter-individually. As it is a common practice in dosimetric studies, we estimated dosimetric data of this model and extrapolated this to our patients.

Although we found that the multiple field RT techniques do not appear to have a major influence on cognitive performance, radiotherapeutic treatment techniques continue to develop to reduce radiation to healthy tissue. Stereotactic RT (SRT) is one of the new RT techniques and is

currently used for the treatment of pituitary tumors in our center. SRT is expected to be a promising alternative treatment to deliver the radiation dose precisely. This technique is expected to spare normal tissue more than the multiple field external beam RT. The first SRT study results are promising according to tumor control and clinical status (including changes in neurological status, such as visual function and endocrinological function) at a median follow-up of 25.5 months [39]. Long-term effects on cognitive functioning need to be awaited. In the (nearby) future, alternative forms can be expected like hippocampal sparing RT or even limbic circuit sparing, neural stem cell sparing, and neural progenitor cell sparing RT [5-7]. This may be especially important for patients with benign tumors, with a long life expectancy like NFA patients.

In conclusion, no dose-response relationship could be established, confirming that current multiple field RT techniques and fractionated radiation dose regimens do not result in major differences in cognitive performance involving the hippocampus and PFC in NFA patients.

Declaration of interest

There is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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References

1 Erridge SC, Conkey DS, Stockton D, Strachan MW, Statham PF, Whittle IR, Grant R, Kerr GR, Gregor A. 2009 *R*adiotherapy for pituitary adenomas: long-term efficacy and toxicity. *Radiotherapy and Oncology: journal of the European Scoiety for the Therapeutic Radiotheapy and Oncology* 93 597-601.

2 van den Bergh AC, van den Berg G, Schoorl MA, SLuiter WJ, van der Vliet AM, Hoving EW, Szabó BG, Langendijk JA, Wolffenbuttel BH, Dullaart RP. 2007 Immediate postoperative radiotehrapy in residual nonfunctioning pituitary adenoma: beneficial effect on local control without additional negative impact on pituitary function and life expectancy. *International Journal of Radiation Oncology, Biology, Physics* 67 863-869.

3 Gittoes NJ. 2005 Pituitary radiotherapy: current controversies. T*rends in Endocrinology and Metabolism* 16 407-413.

4 Lawrence YR, Li XA, el Naqa I, Hahn CA, Marks LB, Merchant TE, Dicker AP. 2010 Radiation dose-volume effects in the brain. *International Journal of Radiation Oncology, Biology, Physics* 76 S20-S27.

5 Gondi V, Tome WA, Mehta MP. 2010 Why avoid the hippocampus? A comprehensive review. *Radiotherapy and Oncology: journal of the European Scoiety for the Therapeutic Radiotherapy and Oncology* 97 370-376.

6 Marsh JC, Gielda BT, Herskovic AM, Abrams RA. 2010 Cognitive Sparing during the Administration of Whole Brain Radiotherapy and Prophylactic Cranial Irradiation: Current Concepts and Approaches. *Journal of Oncology* 198208.

7 Redmond KJ, Achanta P, Grossman SA, Armour M, Reyes J, Kleinberg L, Tryggestad E, Quinones-Hinojosa A, Ford EC. 2011 A radiotherapy technique to limit dose to neural progenitor cell niches without compromising tumor coverage. *Journal of Neurooncology* 104 579-87.

8 Armstrong CL, Hunter JV, Ledakis GE, Cohen B, Tallent EM, Goldstein BH, Tochner Z, Lustig R, Judy KD, Pruitt A, Mollman JE, Stanczak EM, Jo MY, Than TL, Phillips P. 2002 Late cognitive and radiographic changes related to radiotherapy: initial prospective findings. *Neurology* 59 40-48.

9 Armstrong CL, Gyato K, Awadalla AW, Lustig R, Tochner ZA. 2004 A critical review of the clinical effects of therapeutic irradiation damage to the brain: the roots of controversy. *Neuropsychology Review* 14 65-86.

10 Hahn CA, Zhou SM, Raynor R, Tisch A, Light K, Shafman T, Wong T, Kirkpatrick J, Turkington T, Hollis D, Marks LB. 2009 Dose-dependent effects of radiation therapy on cerebral blood flow, metabolism, and neurocognitive dysfunction. *International Journal of Radiation Oncology, Biology, Physics* 73 1082-1087.

11 Surma-aho O, Niemelä M, Vilkki J, Kouri M, Brander A, Salonen O, Paetau A, Kallio M, Pyykkönen J, Jääskeläinen J. 2001 Adverse long-term effects of brain radiotherapy in adult low-grade glioma patients. *Neurology* 56 1285-1290.

12 Noad R, Narayanan KR, Howlett T, Lincoln NB, Page RC. 2004 Evaluation of the effect of radiotherapy for pituitary tumors on cognitive function and quality of life. *Clinical Oncology (Royal College of Radiologists (Great Britain))* 16 233-237.

13 van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, Langendijk JA, Wolffenbuttel BH. 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated for nonfunctioning pituitary adenoma. *International Journal of Radiation Oncology, Biology, Physics* 68 986-991.

14 Guinan EM, Lowy C, Stanhope N, Lewis PD, Kopelman MD. 1998 Cognitive effects of pituitary tumors and their treatments: two case studies and an investigation of 90 patients. *Journal of Neurology, Neurosurgery, and Psychiatry* 65 870-876.

15 McCord MW, Buatti JM, Fennell EM, Mendenhall WM, Marcus RB, Rhoton AL, Grant MB, Friedman WA. 1997 Radiotherapy for pituitary adenoma: long-term outcome and sequelae. *International Journal of Radiation Oncology, Biology, Physics* 39 437-444.

16 Peace KA, Orme SM, Thompson AR, Padayatty S, Ellis AW, Belchetz PE. 1997 Cognitive dysfunction in patients treated for pituitary tumors. *Journal of Clinical and Experimental Neuropsychology* 19 1-6.

17 Peace KA, Orme SM, Padayatty SJ, Godfrey HP, Belchetz PE. 1998 Cognitive dysfunction in patients with pituitary tumor who have been treated with transfrontal or transsphenoidal surgery or medication. *Clinical Endocrinology (Oxf)* 49 391-396.

18 Abayomi OK. 2002 Pathogenesis of cognitive decline following therapeutic irradiation for head and neck tumors. *Acta Oncologica* 41 346-351.

19 Addis DR, Moscovitch M, McAndrews MP. 2007 Consequences of hippocampal damage across the autobiographical memory network in left temporal lobe epilepsy. *Brain* 130 2327-2342.

20 Pereira AG, Portuguez MW, da Costa DI, Azambuja LS, Marroni SP, da Costa JC, Pereira-Filho AA. 2011 Route Learning Performance: Is it a Hippocampus Function? *Cognitive and Behavioral Neurology: official journal of the Society for Behavioral and Cognitive Neurology* 24 4-10.

21 Monje ML, Palmer T. 2003 Radiation injury and neurogenesis. *Current Opinion in Neurology* 16 129-134.

22 Shallice T, Burgess P. 1996 The domain of supervisory processes and temporal organization of behaviour. Philosophical Transactions of the Royal Society of London. Series B Biological Sciences 35 1405-1411.

23 Miller EK, Cohen JD. 2001 An integrative theory of prefrontal cortex function. *Annual Review of Neuroscience* 24 167-202.

24 Brummelman P, Elderson MF, Dullaart RP, van den Bergh AC, Timmer CA, van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van Beek AP. 2011 Cognitive functioning in patients treated for nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Clinical Endocrinology (Oxf)* 74 481-487.

25 Gazzaniga M, Ivry R, Mangun R. 2002 In Cognitive Neuroscience, edn 2, New York: W W Norton & Company, p75, 501.

26 Martin J. 1996 In Neuroanatomy Text and Atlas, edn 2, Stamford, CT: Appleton and Lange.

27 Yushkevich PA, Wang H, Pluta J, Das SR, Craige C, Avants BB, Weiner MW, Mueller S. 2010 Nearly automatic segmentation of hippocampal subfields in in vivo focal T2-weighted MRI. *Neuroimage* 53 1208-1224.

28 van der Elst W, Van Boxtel MP, Van Breukelen GJ, Jolles J. 2005 Rey's verbal learning test: normative data for 1855 healthy participants aged 24-81 years and the influence of age, sex, education, and mode of presentation. *Journal of the International Neuropsychological Society* 11 290-302.

29 Ruff RM, Light RH, Evans RW. 1987 The Ruff Figural Fluency Test: A normative study with adults. *Developmental neuropsychology* 3 37-51.

30 United Nations Educational, Scientific and Cultural Organization. International Standard Classification of Education ISCED. 2006.

31 Spinhoven P, Ormel J, Sloekers PP, Kempen GI, Speckens AE, Van Hemert AM. 1997 A validation study of the Hospital Anxiety and Depression Scale (HADS) in different groups of Dutch subjects. *Psychological Medicine* 27 363-370.

32 Stolk RP, Rosmalen JG, Postma DS, de Boer RA, Navis, G, Slaets JP, Ormel J, Wolffenbuttel BH. 2008 Universal risk factors for multifactorial diseases: LifeLines: a three-generation population-based study. *European Journal of Epidemiology* 23 67-74.

33 Duncan DB. 1955 Multiple range and multiple F tests. *Biometrics* 11 1-42.

34 Tiemensma J, Kokshoorn NE, Biermasz NR , Keijser BJ, Wassenaar MJ, Middelkoop HA, Pereira AM, Romijn JA. 2010 Subtle cognitive impairments in patients with long-term cure of Cushing's disease. *Journal of Clinical Endocrinology and Metabolism* 95 2699-2714.

35 Jalali R, Mallick I, Dutta D, Goswami S, Gupta T, Munshi A, Desphande D, Sarin R. 2010 Factors influencing neurocognitive outcomes in young patients with benign and low-grade brain tumors treated with stereotactic conformal radiotherapy. *International Journal of Radiation Oncology, Biology, Physics* 77 974-979.

36 Douw L, Klein M, Fagel SS, van den Heuvel J, Taphoorn MJ, Aaronson NK, Postma TJ, Vandertop WP, Mooij JJ, Boerman RH, Beute GN, Sluimer JD, Slotman BJ, Reijneveld JC, Heimans JJ. 2009 Cognitive and radiological effects of radiotherapy in patients with low-grade glioma: long-term follow-up. *Lancet Neurology* 8 810-818.

37 Loeffler JS, Shih HA. 2011 Radiation Therapy in the Management of Pituitary Adenomas. *Journal of Clinical Endocrinology and Metabolism* 96 1992-2003.

38 Thaker U, McDonagh AM, Iwatsubo T, Lendon CL, Pickering-Brown SM, Mann DM. 2003 Tau load is associated with apolipoprotein E genotype and the amount of amyloid beta protein, Abeta40, in sporadic and familial Alzheimer's disease. *Neuropathology and Applied Neurobiology* 29 35-44.

39 Hashizume C, Mori Y, Kobayashi T, Shibamoto Y, Nagai A, Hayashi N. 2010 Stereotactic radiotherapy using Novalis for craniopharyngioma adjacent to optic pathways. *Journal of Neurooncology* 98 239-247.

Chapter 4

Cognitive performance and brain abnormalities on MRI in patients treated for nonfunctioning pituitary macroadenoma

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Abstract

Objective The extent to which cognitive dysfunction is related to specific brain abnormalities in patients treated for pituitary macroadenoma is unclear. Therefore, we compared brain abnormalities seen on Magnetic Resonance Imaging (MRI) in patients treated for nonfunctioning pituitary macroadenoma (NFA) with or without impairments in cognitive functioning.

Methods In this cross-sectional design, a cohort of 43 NFA patients was studied at the University Medical Center Groningen. White matter lesions (WMLs), cerebral atrophy, (silent) brain infarcts and abnormalities of the temporal lobes and hippocampi were assessed on pre-treatment and post-treatment MRI scans. Post-treatment cognitive examinations were performed using a verbal memory and executive functioning test. We compared our patient cohort with large reference populations representative of the Dutch population.

Results One or more impairments on both cognitive tests were frequently observed in treated NFA patients. No treatment effects were found with regard to the comparison between patients with and without impairments in executive functioning. Interestingly, in patients with one or more impairments on verbal memory function, treatment with radiotherapy had been given more frequently (74% in the impaired group versus 40% in the unimpaired group, $P = 0.025$). Patients with or without any brain abnormality on MRI did not differ in verbal memory or executive functioning.

Conclusions Brain abnormalities on MRI are not observed more frequently in treated NFA patients with impairments compared to NFA patients without impairments in verbal memory or executive functioning. Conversely, the absence of brain abnormalities on MRI does not exclude impairments in cognition.

Introduction

A nonfunctioning pituitary macroadenoma (NFA) is a benign tumor of the pituitary gland. Its treatment usually consists of pituitary surgery sometimes followed by radiotherapy (RT) in case of incomplete resection or tumor recurrence. Despite the benefits of postoperative RT regarding local control [1,2], concerns related to brain abnormalities and diminished cognitive performance have questioned the safety of RT.

In patients with low grade glioma, white matter lesions (WMLs) and cerebral atrophy are well known radiologically detectable abnormalities of the brain after RT. Furthermore, there appears to be a correlation between radiologic abnormalities and cognitive performance [3,4]. One prospective study found that cognitive performance of irradiated patients with recurrent malignant glioma may be more sensitive in predicting tumor progression than the MR images, because cognitive performance predicted tumor recurrence more than a month earlier than MRI confirmation [5]. Although the aforementioned studies suggest a relation between brain abnormalities and cognitive performance, a shortcoming of many imaging studies in relation to cognition is that they focused on patients without fixed tumor localizations [3-6]. Consequently, different cognitive domains will be affected by the tumor itself leading to heterogeneous results in brain tumor patients. In contrast, NFA patients compared to primary brain tumor patients (1) have a fixed tumor position, (2) are treated with a lower radiation dose and (3) have no exposure of (other) disease- and treatment related factors, such as chemotherapy. This offers a unique opportunity to study cognition and radiological brain abnormalities without these confounding factors.

Although we previously found that NFA patients (tested between September 2008 and December 2009) score significantly worse on cognition compared to reference samples, we did not find a relation between RT and cognitive performance [7,8]. It is, however, possible that brain abnormalities are related to cognitive performance. Up to date, there are no studies on such relationships in patients with a fixed tumor localization within one part of the brain who are treated with relatively low radiation doses. Therefore, the main objective of this follow-up study was to assess and compare cognition and brain abnormalities as shown by MRI in treated (surgery with or without RT) NFA patients with and without cognitive impairments.

Methods

Patients

A total of 43 NFA patients (28 males and 15 females, aged 61 ± 10 years , range 40 – 81 years at time of cognitive assessment) underwent treatment between August 1987 and May 2008, and were evaluated by using cognitive tests and MRI scans at the University Medical Center Groningen (UMCG). All patients were treated with surgery as the primary treatment modality with or without postoperative RT. The diagnosis of NFA was based on the patient's clinical history and presentation, endocrine evaluation, MR imaging, and was confirmed in all patients by postoperative histopathological findings. Pre- and post-treatment MRI scans were available for all patients. The pre-treatment scans were made prior to surgery. To assure that acute and early-delayed post-treatment effects had resolved, patients underwent cognitive testing if surgery or RT was at least

6 months ago and if their hormone replacement schedules were stable during these preceding months. All patients had regular follow-up appointments in our endocrine or radiotherapy outpatient clinic. Patients were followed with MRI scans from the onset of diagnosis until November 2010. Approval was given by the medical ethics review committee of the UMCG.

Cognitive tests

Aspects of verbal memory were assessed with the 15 Words Test (15 WT) which is a Dutch equivalent of the Rey Auditory Verbal Learning Test [9]. In this test, 15 words were presented five times. After each trial, patients were asked to name immediately the words they remembered. This allowed the calculation of three different scores describing *immediate memory*:

1. The *short-term memory* score is based on the number of words patients were able to name after the first presentation of the word list.
2. The *total memory score* represents the total number of words patients remembered over the five trials.
3. The *learning score* describes the difference between the number of words remembered in the third trial in comparison with the first trial.

Besides immediate memory, *delayed memory* was measured.

4. The *delayed memory score* is based on the number of words patients could recall after a period of about 30 minutes.

Executive functioning was assessed using the Ruff Figural Fluency Test (RFFT) [10]. In this test, patients were presented with sheets of paper on which 35 squares were printed, each with a fixed pattern of five dots. The test consisted of five parts, which differed with regard to the designs. While the configurations of dots are the same in the first three parts of the test, two types of distractions are added in two of these parts. In the last two parts the configurations of the dots are different and without distractions. The participant was asked to produce as many different designs as possible by connecting two or more dots in each square with straight lines. The time for each part was restricted to 1 minute so that the total test time was 5 minutes. Responses were scored with regard to the total number of *unique designs* generated over the five parts. The *perseverative errors score* represents the total number of repetitions of the same design drawn. The interrater variability (two independent raters) was determined by Pearson's *r* and was 0.99 for both total unique designs and perseverative errors. The *error ratio* is calculated by the total number of perseverative errors divided by the total number of unique designs.

Questionnaires and protocol

A common questionnaire on demographic and health related data was used with special attention for educational level, social status, full-time/part-time employment, social security benefit, comorbidity, use of medication, cardiovascular risk factors, traumatic brain injury and dementia. Education level was determined by using a Dutch education system, comparable to the International Standard Classification of Education (ISCED) [11]. This scale ranges from 1 (elementary school not finished) to 7 (university level).

The Hospital Anxiety and Depression Scale (HADS) consists of 14 items and measures anxiety and depression [12]. Each item is scored as a number, with a maximum score for each subscale

(anxiety or depression) of 21. Higher scores indicate more severe anxiety or depression.

In fixed order, the test protocol was as follows: (1) the 15 WT: direct recall, (2) the RFFT, (3) a common questionnaire to assess baseline information, (4) physical examination: height, weight, blood pressure, waist circumference, hip circumference and compliance to the test situation (5) the HADS and finally (6) the 15 WT: delayed recall. The assessment took approximately forty minutes and was performed directly after or just before the regular outpatient visit. All testing and scoring of tests were performed by trained personnel.

Reference data: healthy control subjects

The performances of patients were compared with Dutch controls. Normative data for the HADS were derived from Spinhoven *et al.* [12]. In their study, psychometric properties of the HADS were assessed in six different groups of Dutch subjects (n = 6165): (1) a random sample of younger adults (18-65 years) (n = 199); (2) a random sample of elderly subjects of 57 to 65 years of age (n = 1901); (3) a random sample of elderly subjects of 66 years and older (n = 3293); (4) a sample of consecutive general practice patients (n = 112); (5) a sample of consecutive general medical out-patients with unexplained somatic symptoms (n = 169); and (6) a sample of consecutive psychiatric out-patients (n = 491). In all six groups, an authorized Dutch translation of the HADS was used. General population mean and standard deviation scores were used from 18 to 65 years and >65 years to calculate *Z-scores*.

Reference data for the 15 WT were derived from control subjects of the Maastricht Aging Study. In this cohort, 1780 healthy participants between 24 and 81 years were evaluated on a Dutch adaptation of the Rey Verbal Learning Test, the 15 WT [9]. Regressions models given by the authors were used to determine accurate Z-scores. The final test scores were controlled for age, sex, and education. Reference data for the RFFT were derived from a sample (n = 10,289) of the LifeLines Cohort Study [13]. Reference groups were stratified by a matrix of eight education levels and 13 age levels (half decades from 20 to 85 years). Each cluster consisted on average of 120 subjects. RFFT forms were analyzed by a computerized pattern recognition program. There was a good internal consistency (n=373) between computerized rating and human rating for unique designs (Cronbach's $\alpha = 0.99$) and perseverative errors (Cronbach's $\alpha = 0.97$). Using a Bland-Altman analysis, a near perfect level of agreement was found between these two rating methods with intraclass correlation for unique designs of 0.99 and perseverative errors of 0.96. Using the mean and standard deviation scores for each reference group, we standardized our patient scores by converting them into *Z-scores*.

MRI scans

The pre- and post-treatment MRI scans were all obtained from the Department of Radiology at the UMCG from the period July 1987 to November 2010. The MRI scans were primarily done for clinical diagnosis of the NFA and post-treatment follow-up. All patients underwent MRI scanning at the UMCG following a standard protocol. The scans were made on a 1 Tesla system using a regular head coil. The standard scan protocol consisted of coronal T2, and coronal and sagittal T1 of the region of the pituitary gland. On the coronal T2 images the white matter of the frontal and temporal lobes were assessed, as well as the hippocampi and the frontal and temporal horns and

the peripheral cerebrospinal fluid (CSF) spaces. The total number of MRI scans per patient was on average 7 (interquartile range (IQR) 5-8). The median duration of MRI scan follow-up was 6 years (IQR 3-9 years).

Rating of MRI abnormalities

MRI scans were presented randomly and rated by an experienced neuroradiologist (LM) blinded to the clinical data and treatment. The coronal T2 weighted images were used for the assessment of WMLs and central and peripheral CSF spaces (atrophy), (silent) brain infarcts, and any abnormalities of the temporal lobes and left and right hippocampus.

WMLs were defined as subcortical or periventricular focal or confluent areas of abnormal high signal intensity on T2 in the white matter. *Cerebral (central and peripheral) atrophy* was determined by assessing an increase in the peripheral CSF spaces with an increase in the width of the sulci, loss of parenchyma, and dilatation of the lateral and third ventricles. A four point visual rating scale (0-3) was used to assess the severity of the cerebral atrophy (i.e. sulcal and ventricular enlargement): (0) normal, (1) minimal (slightly prominent peripheral CSF spaces, normal ventricles), (2) moderate (prominent peripheral CSF spaces, slightly enlarged ventricles), (3) severe (marked prominence of peripheral CSF spaces, marked increase in size of ventricles) [14]. Cerebral atrophy was rated on T2-weighted MRI scans. *Brain infarcts* (silent and symptomatic) were rated as present or absent and defined as larger areas of T2 hyperintense gliosis or focal tissue loss with the same intensity as cerebrospinal fluid on T1 or T2 weighted MR images. Furthermore, *any MRI abnormalities of the temporal lobes and the size of the both hippocampi* were rated on T2-weighted images as present or absent.

Statistical analysis

All statistical analyses were performed using Statistical Package for the Social Sciences (SPSS, Inc., Armonk, NY, USA), version 20. Demographic data are presented as median, IQR, frequencies or percentages. Since not all data were normally distributed, patients' cognitive performances were compared with the reference group by using non-parametric tests. Cognitive performance data were presented as mean *Z-score* with a non-parametric 95% confidence interval (CI) [15]. Furthermore, the number of patients scoring in the impaired range was presented. According to Lezak, cognitive impairment on a test was defined as a performance equivalent to or below the 10th percentile of the reference samples (equivalent to a *Z-score* of ≤ -1.3) [16]. Categorical variables (e.g. the presence or absence of radiological brain abnormalities) were analyzed using Chi-square test. Continuous variables were analyzed by the Mann-Whitney test. The two-tailed alpha level of <0.05 was considered statistically significant.

Results

Patient characteristics

A total of 43 NFA patients were evaluated for cognitive functioning and brain abnormalities on MRI scans. The clinical characteristics of these patients are shown in Table 1. All patients underwent primary surgery, 36 patients (84%) by transsphenoidal resection and 7 patients (16%) by

craniotomy. Twenty-five patients (58%) were treated with postoperative pituitary RT (with a median time of seven months after initial surgery).

Of the irradiated patients, twenty-three patients (92%) received daily radiation fractions of 1.8 Gray (Gy) and two patients (8%) received daily radiation fractions of 2 Gy. The total radiation dose was: 45 Gy (n = 21, 84%), 46 Gy (n = 2, 8%) or 50.4 Gy (n = 2, 8%). The RT techniques were: two-field (n = 1, 4%), three-field (n = 12, 48%), four-field (n = 3, 12%), five-field (n = 5, 20%), a combination of two-field and three-field (n = 2, 8%) or a combination of two-field and five-field (n = 2, 8%).

Thirty-eight patients (88%) received single or multiple hormone replacement therapy (Table 1).

Cognitive functioning in the whole patient group (n = 43)

Results for cognitive function tests are shown in Table 2. There were no significant differences in *Z-scores* between the irradiated and non-irradiated patients for the verbal memory and executive functioning subscores (data not shown).

Cognitive impairments in patients with or without any brain abnormalities on MRI

Table 3 presents the number of patients with an impaired performance on verbal memory (n = 23, 53%) and executive functioning (n = 19, 44%) divided into a group of patients with no brain abnormalities on MRI versus any brain abnormality on MRI. With regard to the 4 patients with no brain abnormalities on MRI and impaired functioning of verbal memory or executive functioning, there was no overlap in the patients, except for one.

MRI abnormalities in patients with or without cognitive impairments

The characteristics of the MRI abnormalities in NFA patients with or without impairments on verbal memory and executive functioning are given in Table 4 and 5. No differences were seen between both patient groups for WMLs, cerebral atrophy, and abnormalities in temporal lobes or hippocampi. In each group one brain infarct was noted. Also no difference was seen in type of surgery (transsphenoidal surgery or craniotomy) and hormonal substitution therapy for both memory performance and executive functioning. However, patients who received RT had significantly more often impairments in verbal memory compared with patients who had not received RT. In multivariate analyses this finding remained significant when corrected for other treatment characteristics (data not shown).

Table 1. Clinical characteristics of patients treated for a nonfunctioning pituitary adenoma, n = 43

Basic characteristics	
Age at cognitive assessment (y), median [IQR]	60 [56; 70]
Sex (male/female)	28/15
Educational level (1/2/3/4/5/6/7) (number)	1/4/0/9/18/9/2
Surgery (%)	100
Transsphenoidal surgery/ Craniotomy (%)	84/16
Age at surgery (y), median [IQR]	49 [40; 55]
Time between surgery and cognitive test performance (y), median [IQR]	11 [6; 15]
Patients with 2nd surgery (%)	7
Radiotherapy (%)	58
Age at radiotherapy (y), median [IQR]	48 [41; 56]
Time between radiotherapy and cognitive testing (y), median [IQR]	13 [9; 19]
MRI scans	
Age at the last MRI scan (y), median [IQR]	58 [55; 65]
Time between cognitive test performance and last MRI scan (y), median [IQR]	1.3 [0.6; 2.5]
No. of hormonal replacements (0/1/2/3/4/5)	(5/9/8/13/8/0)
Glucocorticoids (% of patients substituted)	63
Thyroid hormone (% of patients substituted)	72
Growth hormone (% of patients substituted)	21
Sex hormone (% of patients substituted)	58
Desmopressin (% of patients substituted)	9

IQR: Interquartile range. Education level was determined by using a Dutch education system, comparable to the International Standard Classification of Education[11]. This scale ranges from 1 (elementary school not finished) to 7 (university level)

Table 2. Cognitive performance of patients treated for nonfunctioning pituitary macroadenoma, n = 43

	Median [IQR]	95% CI of the median	Score ≤ 10 perc. N (%)	*P*-value*
Verbal Memory				
15 Words Test				
Short-term memory	-0.29 [-0.79; 0.08]	-0.63; -0.03	9 (21)	0.03
Total memory	-0.80 [-1.20; -0.30]	-1.12; -0.39	14 (33)	<0.001
Learning score	-0.32 [-0.76; 0.07]	-0.58; 0.11	7 (16)	0.18
Delayed memory	-0.90 [-1.20; -0.10]	-1.04; -0.30	14 (33)	<0.001
Executive functioning				
RFFT				
Unique designs	-0.93 [-1.35; -0.10]	-1.15; -0.32	17 (40)	<0.001
Perseverative errors	-0.71 [-1.06; 0.15]	-0.65; 0.61	8 (19)	0.95
Error ratio	-0.40 [-1.00; 0.21]	-0.59; 0.43	8 (19)	0.76

Cognitive performance data are given as *Z*-scores median and Interquartile range [IQR]
CI: Confidence Interval
**P*-values of the 95% CI compared to the reference data

Table 3. Cognitive impairments in patients with or without any brain abnormalities on MRI.

	No abnormalities on MRI N = 9	Any abnormality on MRI N = 34	*P*-value
Age characteristics			
Age at cognitive assessment (y), median [IQR]	57 [49; 61]	62 [58; 70]	0.092
Age at the last MRI scan (y), median [IQR]	56 [48; 60]	60 [55; 69]	0.159
Cognitive functioning			
Impaired functioning on verbal memory (yes/no) Number (frequency, (%))	4/5 (44/56)	19/15 (56/44)	0.541
Impaired functioning on executive functioning (yes/no) Number (frequency, (%))	4/5 (44/56)	15/19 (44/56)	0.986

Table 4. Comparison of MRI abnormalities between patients with or without impairments on verbal memory.

	Impaired patients N = 23 Number (frequency, (%))	**Unimpaired patients N = 20** Number (frequency, (%))	***P*-value**
Radiological Abnormalities			
White-matter lesions (yes/no)	5/18 (22/78)	9/11 (45/55)	0.104
Cerebral Atrophy (cortical/central/both/no)	3/3/12/5 (13/13/52/22)	2/1/12/5 (10/5/60/25)	0.802
Temporal lobes (abnormal/normal/unknown)	3/19/1 (13/83/4)	6/14/0 (30/70/0)	0.278
Hippocampus (abnormal/normal/unknown)	3/19/1 (13/83/4)	3/17/0 (15/85/0)	0.363
Transsphenoidal surgery/ craniotomy	18/5 (78/22)	18/2 (90/10)	0.298
Radiotherapy (yes/no)	17/6 (74/26)	8/12 (40/60)	0.025
Hormonal replacement			
Glucocorticoids (yes/no)	17/6 (74/26)	14/6 (70/30)	0.775
Thyroid hormone (yes/no)	17/6 (74/26)	13/7 (65/35)	0.526
Growth hormone (yes/no)	6/17 (26/74)	5/15 (25/75)	0.935
Sex hormone (yes/no)	15/8 (65/35)	13/7 (65/35)	0.988
Desmopressin (yes/no)	3/20 (13/87)	1/19 (5/95)	0.365

Table 5. Comparison of MRI abnormalities between patients with or without impairments on executive functioning.

	Impaired patients N = 19 Number (frequency, (%))	**Unimpaired patients N =24** Number (frequency, (%))	***P-value***
Radiological Abnormalities			
White-matter lesions (yes/no)	8/11 (42/58)	6/18 (25/75)	0.235
Cerebral Atrophy (cortical/central/both/no)	2/4/9/4 (11/21/47/21)	3/0/15/6 (13/0/62/25)	0.133
Temporal lobes (abnormal/normal/unknown)	5/14/0 (26/74/0)	4/19/1 (17/79/4)	0.521
Hippocampus (abnormal/normal/unknown)	2/16/1 (11/84/5)	4/20/0 (17/83/0)	0.461
Transsphenoidal surgery/ craniotomy	15/4 (79/21)	21/3 (88/12)	0.451
Radiotherapy (yes/no)	12/7 (63/37)	13/11 (54/46)	0.553
Hormonal replacement			
Glucocorticoids (yes/no)	15/4 (79/21)	16/8 (67/33)	0.373
Thyroid hormone (yes/no)	16/3 (84/16)	14/10 (58/42)	0.067
Growth hormone (yes/no)	6/13 (32/68)	5/19 (21/79)	0.423
Sex hormone (yes/no)	13/6 (68/32)	15/9 (63/37)	0.686
Desmopressin (yes/no)	1/18 (5/95)	3/21 (13/87)	0.417

Discussion

This study confirms that impairments in verbal memory and executive functioning are frequently observed in treated NFA patients. However, patients with and without cognitive impairments did not differ with respect to brain abnormalities on MRI.

To our knowledge, this is the first study that related cognitive functioning to abnormalities in the brain on MRI in a patient group with a fixed position of the tumor. We recruited patients with NFA only, thereby excluding confounding variables such as tumor malignancy and diffuse brain tumor localization which affect different brain regions and hence cognitive functioning. Previously, we found no negative effects of RT in NFA patients on quality of life and objective and self-rated cognitive performance compared to reference populations [7, 17]. These results were extended, by showing an absence of a relationship between cognitive performance and radiation dosimetry [8]. In the present study, 79% of NFA patients had an abnormality on MRI, although these brain abnormalities were not more frequently observed in patients with cognitive impairments. Interestingly, patients who received RT suffered significantly more often from impairments of verbal memory. The differences between our previous findings on the effects of RT and this study can be explained by the fact that a cognitive impairment in the present study was defined as at least one impairment for the total domain of memory, while our previous studies compared impairments for memory subscores [7,8]. However, both studies demonstrate that patients show memory impairments more often compared to the reference population.

In addition to RT, other factors, like surgery, the pituitary disease itself or hormonal abnormalities secondary to the tumor and/or its treatment may lead to impaired cognitive functioning in treated NFA patients [18]. With respect to hormonal abnormalities, glucocorticoids and thyroid substitution therapy are of particular importance for cognitive functioning [19-21]. Glucocorticoids may play an important role in the regulation of memory [19]. In addition to memory, we found that glucocorticoids might also affect other domains of cognition, such as executive functioning, attention and social cognition (P Brummelman, J Koerts, RPF Dullaart, G van den Berg, MM van der Klauw, O Tucha, BHR Wolffenbuttel & AP van Beek, unpublished observations). Regarding thyroid substitution therapy, research in congenital hypothyroid patients or hypothyroidism in adulthood has shown the importance of thyroid hormone for cognition [20, 21]. Although we found no significant differences between these hormonal substitution therapies and impaired performances in memory and executive functioning, a more extensive neuropsychological test battery might provide more information regarding the relation between hormonal substitution therapy and cognitive functioning.

Several limitations need to be addressed. First, the coronal T2 images used for the assessment provide a limited view of the cerebral hemispheres. However, complications which may be associated with surgical and RT treatment for NFA patients would be expected in the frontal and temporal lobes, which are depicted on these images. Second, the patients differed in time interval between cognitive test performance and last MRI scan. However, we found no effects of this time interval on the cognitive outcome measures and the presence or absence of brain abnormalities.

The potential damaging effect of treatment on cognitive functioning is an important factor

in the determination of the risks versus benefits of treatment, and therefore an important part of clinical decision making. Since more than 90% of patients with brain tumors have cognitive impairments at baseline [22], there is need for prospective studies to establish the effects of treatment on cognitive functioning and radiological brain abnormalities. These prospective studies with pre- and post-treatment assessments (MRI scans and cognitive test performance) would inform us about the causal relationship between treatment induced brain damage and cognitive functioning, allowing the discrimination of disease- and treatment related factors.

From this study it is evident that patients can experience complaints in memory and executive functioning which might have vast consequences on the patients' everyday lives (e.g. they find difficulties in managing the day). Clinicians have to be aware that the absence of abnormalities on MRI does not exclude impairments of cognition. Cognitive rehabilitation therapy can be applied to those patients who suffer from cognitive impairments in their daily lives in order to develop compensatory techniques for the management of their lives.

In conclusion, brain abnormalities on MRI are not more frequently observed in treated NFA patients with impairments of verbal memory or executive functioning than in patients without these impairments. Vice versa, the absence of abnormalities on brain MRI does not exclude impairments of cognition.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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References

1 Erridge SC, Conkey DS, Stockton D, Strachan MW, Statham PF, Whittle IR, Grant R, Kerr GR, Gregor A 2009 Radiotherapy for pituitary adenomas: long-term efficacy and toxicity. *Radiotherapy and Oncology: journal of the European Scoiety for the Therapeutic Radiotheapy and Oncology* 93(3): 597-601.

2 van den Bergh AC, van den Berg G, Schoorl MA, SLuiter WJ, van der Vliet AM, Hoving EW, Szabó BG, Langendijk JA, Wolffenbuttel BH, Dullaart RP 2007 Immediate postoperative radiotherapy in residual nonfunctioning pituitary adenoma: beneficial effect on local control without additional negative impact on pituitary function and life expectancy. *International Journal of Radiation Oncology, Biology, Physics* 67(3): 863-869.

3 Postma TJ, Klein M, Verstappen CC, Bromberg JE, Swennen M, Langendijk JA, Taphoorn MJ, Scheltens P, Slotman BJ, van der Ploeg HM, Aaronson NK, Heimans JJ 2002 Radiotherapy-induced cerebral abnormalities in patients with low-grade glioma. *Neurology* 59(1): 121-123.

4 Douw L, Klein M, Fagel SS, van den Heuvel J, Taphoorn MJ, Aaronson NK, Postma TJ, Vandertop WP, Mooij JJ, Boerman RH, Beute GN, Sluimer JD, Slotman BJ, Reijneveld JC, Heimans JJ 2009 Cognitive and radiological effects of radiotherapy in patients with low-grade glioma: Long-term follow-up. *Lancet Neurol* 8(9): 810-818.

5 Meyers CA, Hess KR 2003 Multifaceted end points in brain tumor clinical trials: Cognitive deterioration precedes MRI progression. *Neuro Oncol* 5(2): 89-95.

6 Armstrong CL, Hunter JV, Ledakis GE, Cohen B, Tallent EM, Goldstein BH, Tochner Z, Lustig R, Judy KD, Pruitt A, Mollman JE, Stanczak EM, Jo MY, Than TL, Phillips P 2002 Late cognitive and radiographic changes related to radiotherapy: Initial prospective findings. Neurology 59(1): 40-48.

7 Brummelman P, Elderson MF, Dullaart RP, Van den Berg AC, Timmer CA, Van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van Beek AP 2011 Cognitive functioning in patients treated for nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Clinical Endocrinology* 74(4): 481-487.

8 Brummelman P, Sattler MG, Meiners LC, Elderson MF, Dullaart RP, Van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van Beek AP 2011 Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex. *Eur J Endocrinol* 166(2): 171-179.

9 Van der Elst W, van Boxtel MP, van Breukelen GJ, Jolles J 2005 Rey's verbal learning test: Normative data for 1855 healthy participants aged 24-81 years and the influence of age, sex, education, and mode of presentation J Int Neuropsychol Soc 11(3): 290-302.

10 Ruff RM, Light RH, Evans RW 1987 The Ruff Figural Fluency Test: A normative study with adults. *Developmental neuropsychology* 3: 37-51.

11 United Nations Educational, Scientific and Cultural Organization. International Standard Classification of Education ISCED, 2006.

12 Spinhoven P, Ormel J, Sloekers PP, Kempen GI, Speckens AE, Van Hemert AM 1997 A validation study of the Hospital Anxiety and Depression Scale (HADS) in different groups of Dutch subjects. *Psychological Medicine* 27(2): 363-370. anal

13 Stolk RP, Rosmalen JG, Postma DS, de Boer RA, Navis, G, Slaets JP, Ormel J, Wolffenbuttel BH 2008 Universal risk factors for multifactorial diseases: LifeLines: a three-generation population-based study. *European Journal of Epidemiology* 23(1): 67-74.

14 Armstrong CL, Hunter JV, Hackney D, Shabbout M, Lustig RW, Goldstein B, Werner-Wasik M, Curran WJ jr. 2005 MRI changes due to early-delayed conformal radiotherapy and postsurgical effects in patients with brain tumors. *Int J Radiat Oncol Biol Phys.* 63(1): 56-63.

15 Campbell MJ, Gardner MJ 1988 Calculating confidence intervals for some non-parametric analysis. *British Medical Journal* 296: 1454-1456.

16 Lezak MD, Howieson D, Loring DW. Neuropsychological Assessment, 4 ed. 2004 New York: Oxford University Press.

17 van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, Langendijk JA, Wolffenbuttel BH 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated for nonfunctioning pituitary adenoma. *Int J Radiat Oncol Biol Phys*; 68(4): 986-991.

18 Peace KA, Orme SM, Padayatty SJ, Godfrey HP, Belchetz PE 1998 Cognitive dysfunction in patients with pituitary tumour who have been treated with transfrontal or transsphenoidal surgery or medication. *Clin Endocrinol (Oxf)* 49(3): 391-6.

19 de Quervain DJ, Aerni A, Schelling G, Roozendaal B 2009 Glucocorticoids and the regulation of memory in health and disease. *Front Neuroendocrinol* 30(3): 358-70.

20 Morreale de Escobar G, Obregon MJ, Escobar del Rey F 2004 Role of thyroid hormone during early brain development. *Eur J Endocrinol* 151 Suppl 3: U25-U37.

21 Osterweil D, Syndulko K, Cohen SN, Pettler-Jennings PD, Hershman JM, Cummings JL, Tourtellotte WW, Solomon DH 1992 Cognitive function in non-demented older adults with hypothyroidism. *J Am Geriatr Soc* 40(4): 325-35.

22 Tucha O, Smely C, Preier M & Lange KW 2000 Cognitive deficits before treatment among patients with brain tumors. *Neurosurgery* 47(2): 324-334.

Hormonal and medical therapy and the effects on cognition

Chapter 5

Effects of previous growth hormone excess and current medical treatment for acromegaly on cognition

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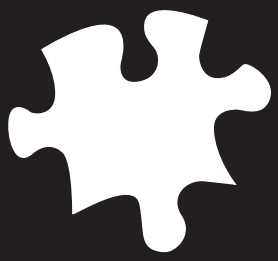

Abstract

Background In untreated acromegaly patients, decreased cognitive functioning is reported to be associated with the degree of growth hormone (GH) and IGF-1 excess. Whether previous GH excess or current medical treatment for acromegaly specifically affects cognition remains unclear. The aim of this study was to compare cognitive functioning of patients who are treated for acromegaly with patients with nonfunctioning pituitary adenomas (NFA). In addition, we assessed the influence of prolonged medical treatment after initial transsphenoidal surgery on cognition.

Design In this cross-sectional study, 74 patients participated, who were treated for acromegaly (n = 50; median [interquartile range] age: 53 [45-65] years) or NFA (n = 24; age: 63 [59-70] years). The NFA patients were selected for a high likelihood of normal GH secretion based on an IGF-1 *Z-score* within the normal range (> -2) and zero or one axis substituted. Of the acromegaly patients, 28 had achieved remission, while 22 were biochemically controlled with long-acting somatostatin analogues and/or pegvisomant. Memory and executive functioning were assessed by the 15 Words Test and the Ruff Figural Fluency Test, and reported as *Z-scores*.

Results The total patient group scored significantly poorer than the reference population on memory and executive functioning ($P < 0.001$). However, cognitive test performance was not significantly different between acromegaly patients with a persistent disease, acromegaly patients in remission and NFA patients.

Conclusion The total patient group scored worse compared with reference populations. We found no association between previous GH excess and cognition. In addition, current medical treatment for GH excess in acromegaly was not related to memory and executive functioning.

Introduction

Changes in cognitive functioning can be expected as a consequence of growth hormone (GH) excess, or GH deficiency. The reason for this assumption is a wide distribution of binding sites for Insulin-like growth factor-1 (IGF-1) in the brain in particular in the medial temporal lobe (the hippocampus) and in the prefrontal cortex (PFC) [1]. Indeed, studies on the effects of GH and IGF-1 on cognition performed in GH deficient patients demonstrated a link between GH and cognitive performance, where poor performance was ameliorated with GH treatment [2-4].

In acromegaly, which is characterized by excess GH production, cognitive functioning is also reported to be impaired. In a recent study, Leon-Carrion *et al.* [5] compared patients with active acromegaly with healthy participants. They found a moderate to severe memory impairment in patients and postulated that high levels of GH and IGF-I in acromegaly patients could be the basis for these findings. However, comparisons with untreated acromegaly patients are less informative because nonspecific psychological factors associated with chronic illness will undoubtedly influence the results [6]. In another study, Sievers and colleagues compared acromegaly patients (of which 60% was either cured or biochemically controlled) with healthy individuals [7]. They found that cognitive dysfunction, particularly attentional deficits, were common in acromegaly. Although this finding is of importance, again comparisons were made with healthy controls. More informative would be a comparison with patients who have nonfunctioning pituitary adenomas (NFA). These patients share many disease characteristics but do not have the GH excess. This type of study was performed by Tiemensma *et al*. [8], who concluded that there was normal cognitive functioning after long-term cure for acromegaly. The relation of previous GH excess with impaired cognitive functioning remains however controversial due to these inconsistent study results.

In acromegaly, GH and IGF-1 may be discordant on medication during persistent disease. Biermasz *et al.* [9] demonstrated that octreotide did not lead to orderly GH secretion in acromegaly in spite of normalization of IGF-1. Neggers *et al.* [10] postulated that long-acting somatostatin analogues normalize serum IGF-1 levels in certain patients in the presence of elevated GH actions in extrahepatic tissues. This phenomenon was called extrahepatic acromegaly. This change in diurnal GH profile, while IGF-1 is normalized, potentially also influences cognitive functioning. Until now no research is available on the effects of medical treatment in acromegaly on cognition.

With these considerations in mind, we studied acromegaly patients with persistent disease (i.e. on GH suppressive mediation) and compared them with acromegaly patients who were in remission. In addition, comparisons were made to NFA patients, to investigate the effects of previous GH excess.

Methods and materials

Patients

In this cross-sectional study, patients were recruited for participation at the endocrine outpatient clinic of the University Medical Center Groningen (UMCG), a tertiary referral center for pituitary surgery in the Netherlands. Inclusion criteria were as follows: age ≥ 18 years, treatment for acromegaly or NFA and regular follow-up in our endocrine outpatient clinic (i.e. at least once a year).

Additional in- and exclusion criteria, details of the study protocol and definitions of hormone deficiencies are described elsewhere [11]. Briefly, NFA and acromegaly patients were tested between September 2008 and November 2011. The diagnosis of NFA was based on two criteria: presence of a pituitary macroadenoma (>1 cm) on magnetic resonance imaging and absence of overproduction of any of the pituitary hormones. The selection of the NFA patients was based on the absence of GH substitution, IGF-1 *Z-scores* within the age-adjusted normal range (> - 2) and zero or one axis being substituted. Pituitary deficiencies were defined according to generally accepted guidelines. The initial diagnosis of acromegaly was based on the characteristic clinical signs and symptoms and confirmed by insufficient suppression of GH during an oral glucose tolerance test (oGTT), elevated age-adjusted IGF-1 concentrations and the presence of a pituitary adenoma on radiologic imaging. Of the acromegaly patients who were included in this study, 28 were in remission (i.e. they had IGF-1 levels in the normal range, and a normal suppression of GH on oGTT (< 1.0 µg/L)), while 22 had persistent disease and were still treated with long-acting somatostatin analogues and/or pegvisomant. The consecutive approach to our acromegaly study patients was considered to be representative for the patient group, as published previously for NFA patients [12].

IGF-1 assay

Plasma IGF-1 was measured by radioimmunoassay after acid-ethanol extraction (Nichols Institute of Diagnostics, San-Juan Capistrano, CA, USA) [13]. IGF-1 was measured on the day of testing. Data are reported as IGF-1 *Z-scores*.

Cognitive tests

Aspects of verbal memory were assessed with the 15 Words Test (15 WT) which is a Dutch equivalent of the Rey Auditory Verbal Learning Test [14]. In this test, 15 words were presented five times. After each trial, patients were asked to name immediately the words they remembered. This allowed the calculation of three different scores describing *immediate memory*:

1. The *short-term memory score* is based on the number of words patients were able to name after the first presentation of the word list.
2. The *total memory score* represents the total number of words patients remembered over the five trials.
3. The *learning score* describes the difference between the number of words remembered in the third trial in comparison with the first trial.

Besides immediate memory, *delayed memory* was measured.

4. The *delayed memory score* is based on the number of words patients could recall after a period of about 30 minutes.

Executive functioning was assessed using the Ruff Figural Fluency Test (RFFT) [15]. In this test, patients were presented with sheets of paper on which 35 squares were printed, each with a fixed pattern of five dots. The test consisted of five parts, which differed with regard to the designs. While the configurations of dots are the same in the first three parts of the test, two types of distractions are added in two of these parts. In the last two parts, the configurations of the dots are different and without distractions. The participant was asked to produce as many different designs as possible by connecting two or more dots in each square with straight lines. The time for each part was restricted to 1 minute so that the total test time was 5 minutes. Responses

were scored with regard to the total number of *unique designs* generated over the five parts. The *perseverative errors score* represents the total number of repetitions of the same design drawn. The interrater variability (two independent raters) was determined by Pearson's *r* and was 0.99 for both total unique designs and perseverative errors. The *error ratio* is the total number of perseverative errors divided by the total number of unique designs.

Questionnaires and protocol

A common questionnaire on demographic and health-related data was used with special attention for educational level, social status, full-time/part-time employment, social security benefit, comorbidity, use of medicine, cardiovascular risk factors, traumatic brain injury and dementia. Education level was determined using a Dutch education system, comparable to the ISCED [16]. This scale ranges from 1 (elementary school not finished) to 7 (university level).

The Hospital Anxiety and Depression Scale (HADS) was used to measure symptoms of anxiety and depression and consists of 14 items [17]. Each item is scored as a number, with a maximum score for each subscale (anxiety or depression) of 21. Higher scores indicate more severe anxiety or depression.

In fixed order, the test protocol was as follows: (1) the 15 WT: direct recall, (2) the RFFT, (3) a common questionnaire to assess baseline information, (4) physical examination: length, weight, blood pressure, waist circumference, hip circumference and compliance to the test situation, (5) the HADS and finally (6) the 15 WT: delayed recall.

The assessment took approximately forty minutes and was performed directly after or just before patients' visit to the outpatient clinic. All testing and scoring of tests were performed by trained personnel.

Reference data: healthy control subjects

The performances of patients were compared to Dutch controls. Normative data for the HADS were derived from Spinhoven *et al* [17]. In this study, general population mean and standard deviation scores were used from 18 to 65 years and >65 years to calculate *Z-scores*. Reference data for the 15 WT were derived from control subjects of the Maastricht Aging Study (n = 1780, aged 24 to 81 years) [14]. Regressions models given by the authors were used to determine accurate *Z-scores*. The final test scores were controlled for age, sex, and education. Reference data for the RFFT were derived from a sample (n = 10,289, half decades from 20 to 85 years) of the LifeLines Cohort Study [18]. Using the mean and standard deviation scores from this cohort, we standardized our patient scores by converting it into *Z-scores*.

Statistical analyses

The analyses were all carried out using the Predictive Analytics SoftWare (PASW; SPSS, Inc., Armonk, NY, USA) statistics 18 package. Demographic data are presented as median and interquartile range (IQR), frequencies or percentages. Normality of data was analyzed by QQ-plots. Not all data were distributed normally; therefore, a nonparametric method was used to test for differences between the groups on the cognitive measures. Categorical variables were analyzed using Chi-square tests. Continuous variables were analyzed by the Kruskal-Wallis ANOVA.

Cognitive performance data were presented as median and IQR. A nonparametric 95% confidence interval (CI) was calculated according to the method of Campbell and Gardner [19]. Furthermore, an impaired performance was defined as a score ≤ 1.3 SD below the normative mean. Only 10% of the overall normal population would be expected to score in this range [20]. We compared three groups: acromegaly patients with persistent disease, acromegaly patients in remission and NFA patients. The two-tailed alpha level of <0.05 was considered statistically significant. In case of statistical differences between the groups on demographic or cognitive data, a Bonferroni correction was performed to protect against Type I error when performing multiple comparisons. In case of statistical differences between the groups on demographic or cognitive data, Duncan's method was used as a *post-hoc* test [21].

Reporting of the study conforms to STROBE statement along with references to STROBE statement and the broader EQUATOR guidelines [22].

Results

Patients

Seventy-four patients participated in the present study (35 men, 39 women, aged 57 ± 12 years, range 22-81 years). Fifty patients with acromegaly and 24 patients with NFA participated. The acromegaly patient group consisted of 22 patients with a persistent disease and 28 patients who were in remission. Patients' characteristics are given in Table 1. The acromegaly patient groups were significantly younger at surgery, radiotherapy and time of testing compared with the NFA patient group. Acromegaly patients with persistent disease activity received significantly more often radiotherapy compared with acromegaly patients who were in remission and NFA patients. Furthermore, time since radiotherapy was longer in acromegaly patients who were in remission compared with acromegaly patients with persistent disease activity and NFA patients. The IGF-1 *Z-scores* were comparable between both acromegaly groups and significantly higher compared with the NFA patient group, despite highly similar hormonal replacements. Both acromegaly patient groups had significantly higher anxiety *Z-scores* (indicating more anxiety) in comparison with the NFA patient group, although there was no indication for clinical anxiety (*Z-scores* within the normal range).

Table 1 Clinical characteristics of acromegaly patients with persistent disease, acromegaly patients in remission and NFA patients

	Acromegaly		NFA	
	Persistent disease	Patients in remission		***P*-value**
N	22	28	24	
Basic characteristics				
Age (y) Median [IQR]	54 [44; 62]	53 [45; 67]	63 [59; 70]	0.008
Sex (males/females)	11/11	12/16	12/12	0.837
Educational level (1/2/3/4/5/6/7)	0/2/0/4/7/7/2	0/2/0/6/12/8/0	0/1/0/5/10/8/0	0.674
Surgery				
Age at surgery (y) Median [IQR]	47 [38; 56]	43 [36; 55]	58 [51; 64]	0.002
Average time since surgery (y) Median [IQR]	5 [3; 7]	8 [4; 14]	5 [3; 9]	0.203
<1-5 years (number (%))	50	36	50	0.458
5-10 years	36	25	37	
> 10 years	14	39	13	
Patients with 2nd surgery (%)	18	4	0	0.034
Use of GH suppressive medication				
SA (N) (%)	9 (41%)	NA	NA	
Peg (N) (%)	6 (27%)	NA	NA	
SA and Peg (N) (%)	7 (32%)	NA	NA	
Radiotherapy (RT)				
RT after surgery (%)	64	21	17	0.001
Age at RT (y) Median [IQR]	37 [32; 47]	47 [38; 57]	62 [49; 75]	0.046
Average time since RT (y) Median [IQR]	4 [2; 7]	16 [11; 24]	4 [2; 4]	0.004
Hormonal substitution				
No. of hormone replacements (0/1/2/3/4/5)	12/5/3/1/1/0	19/6/2/0/1/0	10/14/0/0/0/0	0.070
Glucocorticoid (%)	32	14	17	0.269
Thyroid hormone (%)	27	14	13	0.356
Growth hormone (%)	0	7	0	0.379
Sex hormone (%)	18	11	29	0.237
Desmopressin (%)	5	4	0	0.579
IGF-1 Z-score Median [IQR]	0.42 [-0.43; 0.60]	0.69 [-0.27; 1.17]	-1.12 [-1.56; -0.52]	<0.001
HADS Z-score				
Anxiety	0.03 (-0.31; 0.54)	0.25 (-0.03; 0.81)	-0.53 (-1.03; 0.40)	0.038
Depression	-1.14 (-0.73; 0.56)	-0.14 (-0.93; 0.94)	-0.44 (-1.00; 1.04)	0.372

NFA: nonfunctioning pituitary adenomas; GH: growth hormone; SA: Somatostatin analogues; Peg: Pegvisomant; HADS: Hospital Anxiety and Depression Scale; IQR: Interquartile range

Cognitive Tests

Cognitive performance data of the three groups are given in Table 2. The 95% CIs fall below zero for all memory scores and for the unique designs of the executive functioning test. For the same variables, a higher proportion of patients scored in the impaired range (all $P < 0.001$).

Z-scores for cognitive functioning in acromegaly patients with persistent disease, acromegaly patients in remission and matched NFA patients are given in Table 3. No significant differences were found in median (IQR) cognitive *Z-scores* between the three groups.

Table 2. Cognitive performance of the total patient group

	Median [IQR]	**95% CI of the median**	**Score ≤ 10 perc. *n***	***P-value****
Memory performance				
15 words test				
Short-term memory	-0.36 [-1.29; 0.45]	-0.61; -0.07	18	<0.001
Total memory	-0.80 [-1.60; 0.33]	-1.00; -0.40	24	<0.001
Learning score	-0.51 [-1.28; 0.53]	-0.74; -0.03	18	<0.001
Delayed memory	-0.80 [-1.73; 0.03]	-1.20; -0.30	27	<0.001
Executive functioning				
Ruff Figural Fluency Test				
Unique designs	-0.52 [-1.51; 0.21]	-0.94; -0.10	22	<0.001
Perseverative errors	1.02 [0.04; 1.27]	0.76; 1.17	5	0.778
Error ratio	0.73 [0.17; 1.14]	0.61; 0.93	6	0.260

CI, Confidence interval; IQR, interquartile range

**P*-values by chi-square test comparing the number of patients who score in the impaired range to the number of patients who scored in the unimpaired range.

Table 3. Cognitive performance of acromegaly patients with persistent disease, acromegaly patients in remission and NFA patients

	Acromegaly		**NFA**	***P*-value**
	Persistent disease	Patients in remission		
	N = 22	N = 28	N = 24	
Memory performance				
15 Words Test				
Short-term memory	-0.74 [-1.69; 0.18]	-0.28 [-0.89; 0.28]	-0.28 [-1.17; 0.61]	0.274
Total memory	-0.85 [-1.63; 0.23]	-0.90 [-1.53; -0.10]	-0.55 [-1.75; 0.60]	0.557
Learning score	-0.03 [-0.65; 1.14]	-0.63 [-1.60; 0.36]	-0.67 [-1.12; 0.48]	0.088
Delayed memory	-0.55 [-1.83; 0.28]	-0.90 [-1.93; -0.10]	0.65 [-1.55; 0.08]	0.714
Executive functioning				
Ruff Figural Fluency Test				
Unique designs	-0.83 [-1.51; 0.00]	-0.29 [-1.42; 0.74]	-0.62 [-1.61; 0.15]	0.538
Perseverative errors	1.05 [-0.32; 1.35]	1.04 [0.04; 1.31]	0.92 [0.26; 1.26]	0.926
Error ratio	0.56 [-0.90; 1.22]	0.80 [0.28; 1.13]	0.67 [0.22; 1.12]	0.691

Cognitive performance data are given as z-scores median [Interquartile range].

NFA: nonfunctioning pituitary adenomas

Discussion

In the present study, we found no association between previous GH excess and cognition. In addition, current medical treatment for GH excess was not related to impairments in memory and executive functioning. Although no differences were found between patient groups, most test results differed significantly from the reference populations, indicating a poor cognitive test performance in both patients with acromegaly and NFA.

Only a few studies have addressed cognition in acromegaly. Sievers *et al*. studied 55 patients with biochemically confirmed acromegaly and compared their data with 87 control subjects. These patients were tested for attention, memory and executive functioning. They found that 33% of the patients had an impaired score (< 16th percentile) in the domain of attention, 24% in the domain of memory and up to 17% in the executive functioning domain. However, no associations between the patient's performance and their status of biochemical control, the current therapeutic regime, or the estimated total time of hormone excess, were found [7]. With our more stringent cut-off values (≤ 10th percentile), we also found a high proportion of patients (72%) scoring in the impaired range on at least one outcome variable independent of their status of biochemical control and the current therapeutic regimen. In a study by Leon-Carrion, moderate to severe memory impairment was found in acromegaly patients compared with healthy controls with decreased neural activity in specific brain areas on quantitative electroencephalogram [5]. A different approach to demonstrate altered cognitive function was applied by Tanriverdi *et al*. [23]. In electrophysiological studies they used the P300 amplitude, which is related to updating of working memory (decision making and memory processing), and found that the mean amplitude at all electrode sites was significantly lower in 18 patients with acromegaly when compared with 16 healthy controls and GH deficient patients. However, the acromegaly patients were enrolled into the study before any medical and surgical intervention. Further, as the authors already acknowledged themselves, the skull thickness may have interfered in the measurements of the P300 amplitudes in acromegaly patients.

We decided to compare patients with biochemical control of acromegaly to patients with a pituitary disease but without previous hormone excess. In accordance with the present findings, Tiemensma *et al*. found that cognitive function in patients cured from acromegaly did not differ from patients treated for NFA. Intriguingly and in contrast to a bulk of evidence indicating that any disease (treatment) with its long-term effects results in impairment of cognitive functioning, they found no differences with matched controls [8].

In our study, all but one of the participating patients had normal IGF-1 *Z-scores*. In spite of this metabolic control, acromegaly patients with persistent disease who are on medication are reported to have a disorderly GH secretion with repressed secretory burst-mass and non-pulsatile secretion on octreotide [9]. Another form of discordance is also described between GH and IGF-1 in patients with acromegaly. Neggers *et al*. [10] postulate that long-acting somatostatin analogues normalize serum IGF-1 levels in certain patients in the presence of elevated GH actions in extrahepatic tissues. For these reasons, it is very interesting to study the effects of medication in patients with persistent disease and compare them with acromegaly patients in remission. Previously, we demonstrated that quality of life is impaired in association with the need for prolonged postoper-

ative therapy by somatostatin analogues in patients with acromegaly [24]. With regard to cognition, Sonino found no significant improvements in cognition after treatment with octreotide [25] while others found improved cognitive functions [26]. However, both studies assessed self-reported cognition which is known to be often unrelated to objective measures of cognition [27-29]. Further, within person comparisons were made during active disease and controlled disease, indicating that the effects may not *per se* be related to GH and IGF-1 concentrations. We found no differences between patients with persistent disease and patients who were in remission in spite of far more extensive treatment in persistent disease. This finding suggests that disease control by normalization of IGF-1 alone might be sufficient for improvements in cognitive functioning. This interpretation however remains to be confirmed because our study is cross-sectional in design and therefore limited to infer causality. Additional studies should take GH secretory profiles in these patients into account.

Some limitations need to be addressed. First, we did not use an extensive battery of cognitive tests as is common in many studies on cognitive functioning. We specifically chose for this design to avoid multiple comparisons and thereby potentially inducing type 1 errors. It allowed us also to test a larger group than generally reported in the literature, thereby enhancing our statistical power. However, for future studies an extensive neuropsychological test battery is recommended, where multiple cognitive domains are evaluated. Secondly, baseline and treatment characteristics were at some points different between groups. Although this is an inherent weakness of all observational studies, potential effects have to be taken into account. Previously, we found no negative effects of radiotherapy on quality of life and cognition [11-13]. In regression analysis, we also found no effects of radiotherapy (data not shown).

In conclusion, we found no effects of previous GH excess on cognition. Additionally, acromegaly patients on GH suppressive medication with normal IGF-1 *Z-scores* performed similarly to acromegaly patients who were in remission and NFA patients despite potential differences in diurnal GH concentrations. Based on these findings, the endocrinologist can inform acromegaly patients that memory and executive functioning are not expected to change as a consequence of the GH excess in history or the GH suppressive medication. However, patient groups had poorer cognitive performance corrected for age, and educational level than the reference population, indicating that other disease related factors may potentially be important for cognition. Therefore, more research is needed in the field of cognition in patients with hormonal deficits.

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References

1 Van Dam PS, Aleman A. 2004 Insulin-like growth factor-I, cognition and brain aging. *European Journal of Pharmacology*; 490: 87-95.

2 Sathiavageeswaran M, Burman P, Lawrence D, Harris AG, Falleti MG, Maruff P. 2007 Effects of GH on cognitive function in elderly patients with adult-onset GH deficiency: a placebo-controlled 12-month study. *European Journal of Endocrinology*; 156: 439-447.

3 Oertel H, Schneider HJ, Stalla K, Holsboer F, Zihl J. 2004 The effect of growth hormone substitution on cognitive performance in adult patients with hypopituitarism. *Psychoneuroendocrinology*; 29: 839-850.

4 Faletti MG, Maruff P, Burman P, Harris A. 2006 The effects of growth hormone (GH) deficiency and GH replacement on cognitive performance in adults: a meta analysis of the current literature. *Psychoneuroendocrinology* 31: 681-691.

5 Leon-Carrion J, Martin-Rodriguez JF, Madrazo-Atutxa A, Soto-Moreno A, Venegas-Moreno A, Torres-Vela E, *et al.* 2010. Evidence of cognitive and neurophysiological impairment in patients with untreated naive acromegaly *J Clin Endocrinolo Metab* 2010; 95: 4367-4379.

6 Van den Berg E, Kloppenburg RP, Kessels RP, Kappelle LJ, Biesels GJ. 2009 Type II diabetes mellitus. hypertension. dyslipidemia and obesity: A systematic comparison of their impact on cognition. *Biochimica et biophysica acta*; 1792: 470-81.

7 Sievers C, Sämann PG, Pfister H, Dimopoulou C, Czisch M, Roemmler J, *et al.* 2012 Cognitive function in acromegaly: description and brain volumetric correlates *Pituitary*; 15:350-7.

8 Tiemensma J, Biermasz NR, Van der Mast RC, Wassenaar MJE, Middelkoop HAM., Pereira AM, *et al.* 2010 Increased psychopathology and maladaptive personality traits, but normal cognitive functioning in patients after long term cure of acromegaly *J Clin Endocrinolo Metab*; 95: E392-E402.

9 Biermasz NR, Pereira AM, Frölich M, Romijn JA, Veldhuis JD, Roelfsema F. 2004 Ocreotide represses secretory-burst mass and nonpulsatile secretion but does not restore event frequency or orderly GH secretion in acromegaly Am *J Physiol Endocrinol Metab*; 286: E25-E30.

10 Neggers SJ, Kopchick JJ, Jørgensen, van der Lely AJ 2011 Hypothesis: Extra-hepatic acromegaly: a new paradigm? *European Journal of Endocrinology*; 164: 11-16.

11 Brummelman P, Sattler MG, Meiners LC, Elderson MF, Dullaart RP, Van den Berg G, *et al.* 2011 Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex. *European Journal of Endocrinology*; 166: 171-9.

12 Brummelman P, Elderson MF, Dullaart RPF., Van den Berg ACM., Timmer CA, Van den Berg G, *et al.* 2011 Cognitive functioning in patients treated for nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Clinical Endocrinology*; 74, 481-487.

13 Van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, Langendijk JA, *et al.* 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated for nonfunctioning pituitary adenoma. *Int J Radiat Oncol Biol Phys*; 68: 986-991.

14 Van der Elst W, Van Boxtel MPJ, Van Breukelen GJP, Jolles J. 2005 Rey's Verbal learning test: Normative data for 1855 healthy participants aged 24-81 years and the influence of age, sex, education, and mode of presentation. *Journal of the International Neuropsychological Society*; 11: 290-302.

15 Ruff RM., Light RH, Evans RW. 1987 The Ruff Figural Fluency Test: A normative study with adults. *Developmental neuropsychology*; 3: 37-51.

16 United Nations Educational. Scientific and Cultural Organization. International Standard Classifications of Education ISCED. 2006.

17 Spinhoven P, Ormel J, Sloekers PP, Kempen GI, Speckens AE, Van Hemert AM 1997 A validation study of the Hospital Anxiety and Depression Scale (HADS) in different groups of Dutch subjects. *Psychological Medicine*; 27 363-370.

18 Stolk RP, Rosmalen JG, Postma DS, de Boer RA, Navis G, Slaets JP, 2008 Universal risk factors for multifactorial diseases: LifeLines: a three-generation population-based study. *European Journal of Epidemiology*; 23 67-74.

19 Campbell MJ, Gardner MJ. 1988 Calculating confidence intervals for some non-parametric analysis British *Medical Journal*; 296: 1454- 1456.

20 Lezak MD, Havieson DB, Loring DW. , Hannay HJ, Fisher JS. Neuropsychological Assessment, New York: Oxford University Press; 2004.

21 Duncan DB, Multiple range and Multiple F tests. *Biometrics*, 1995; 11: 1-42.

22 Simera I, Moher D, Hoey J, Schulz KF, Altman DG. 2010 A catalogue of reporting guidelines for health research. *Eur J Clin Invest* 40: 35-53.

23 Tanriverdi F, Yapislar H, Karaca Z, Unluhizarci K, Suer C, Kelestimur F. 2009 Evaluation of cognitive performance by using P300 auditory event related potentials (ERPs) in patients with growth hormone (GH) deficiency and acromegaly. *GH & IGF-1 research*; 19: 24-30

24 Postma MR, Netea-Maier RT, van den Berg G, Homan J, Sluiter WJ, Wagenmakers MA. 2012 Quality of life is imaired in association with the need for prolonged postoperative therapy by somatostatin analogs in patients with acromegaly. *European Journal of Endocrinology*; 166 585-92.

25 Sonino N, Scarpa E, Paoltta A, Fallo F, Boscaro M. 1999 Slow-release Lanreotide Treatment in Acromegaly: effects of Quality of Life *Psychotherapy and Psychosomatics*; 68: 165-167.

26 Ruchala M, Stangierska I, Gurgul E, Stangierski A, Fajfer J, Sowinski J. 2010 The effect of ocreotide treatment on somatic and psychological symptoms of acromegaly *Neuroendocrinology Letters*; 31; 265-269.

27 Maor Y, Olmer L, & Mozes B 2001 The relation between objective and subjective impairment in cognitive function among multiple sclerosis patients- the role of depression. *Multiple sclerosis*; 7: 131-135.

28 Middleton LS, Denney DR, Lynch SG, Parmenter B. 2006 The relationship between perceived and objective cognitive functioning in multiple sclerosis. Archives of Clinical Neuropsychology; 21: 487-494.

29 Sawrie SM, Martin RC, Kuzniecky R, Faught E, Morawetz R, Jamil F. 1999 Subjective versus objective memory change after temporal lobe epilepsy surgery. *Neurology*; 53: 1511-1517.

Chapter 6

Cognitive functioning in pituitary patients treated for adrenal insufficiency

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Abstract

Background Extensive evidence in healthy volunteers links cognition to plasma glucocorticoid levels. However, no studies have been performed in patients with secondary adrenal insufficiency (SAI) on replacement doses with hydrocortisone aiming to mimic normal physiology. The present study focused on the effects of SAI on cognition by applying a comprehensive cognitive test battery consisting of tests measuring various aspects of memory, attention, executive functioning as well as social cognition.

Methods Sixty patients with SAI participated in the present study (35 men and 25 women, mean(SD) age 52(14) years, range 19-73). Patients were treated with hydrocortisone at the time of assessment (mean(SD) dose, 25(5) mg hydrocortisone). Patients' performances on standardized cognitive tests were compared to age-, sex- and education-matched normative data collected on the healthy population.

Results Data analysis revealed significant impairments of patients in the domains of verbal memory, attention, executive functions and social cognition, while no dysfunctions were found with regard to visual memory. Various treatment modalities and treatment related characteristics were related to impairments in different cognitive domains.

Discussion The cognitive profile indicates that pituitary patients treated for adrenal insufficiency do not show a general cognitive impairment but rather a pattern of selective deficits with some cognitive domains being impaired while others remain preserved. In addition, treatment of pituitary disease in its various forms may affect different cognitive domains.

Introduction

Patients with secondary adrenal insufficiency (SAI) have impaired adrenocorticotropic hormone (ACTH) production and are treated with glucocorticoids (GC) to compensate for the loss of endogenous cortisol production. Usually this is done by oral administration of hydrocortisone (HC) or cortisone acetate (CA) aiming to mimic a normal circadian cortisol rhythm, with peak values in the early morning and low concentrations at bedtime.

GC substitution therapy, despite being referred to as a physiological replacement of GC, has its imperfections. Current GC dose-regimens inevitably result in over- or under-replacement during certain periods of the day. This may reduce quality of life and lead to higher risks of mortality and morbidity in patients with hypopituitarism compared to the normal population [1,2].

Besides a poor quality of life and physical side effects, cognitive side effects have also been reported in healthy individuals treated with GC. There is compelling evidence of an inverted "U"-shape relation between plasma GC levels and cognitive function [3]. In particular, deficits in memory [4-11] and executive functioning [12, 13] are reported in association with higher cortisol doses in healthy volunteers. These impairments could be explained by the fact that memory and executive functioning rely on brain structures containing high concentrations of GC receptors, like the hippocampus [14] and the prefrontal cortex [15]. Indeed, chronically elevated cortisol levels are associated with a reduced hippocampal volume and impairments in memory [8]. The prefrontal cortex is, however, not only associated with executive functioning, but also with social cognition [16] and attention [17].

To our knowledge there are no studies examining the effects of long-term HC replacement on cognition in patients with SAI. Therefore, the aim of this study was to examine cognitive functioning with a comprehensive cognitive test battery in pituitary patients treated for adrenal insufficiency.

Methods

Patients

In this cross-sectional study, patients were recruited for participation at the endocrine outpatient clinic of the University Medical Center Groningen (UMCG), a tertiary referral center for patients with pituitary diseases in the Netherlands. A total of 60 patients participated in this study. All patients had SAI for which they received GC substitution therapy. To avoid effects of different types of GC, all patients on CA were converted to treatment with HC in a bioequivalent dose, (i.e. CA dose (in mg) times 0.8 when compared to HC dose (mg)) during a four week run-in phase. The diagnosis of SAI was based on internationally accepted biochemical criteria, principally early morning (08.00 – 09.00 am) serum cortisol measurements and if necessary an insulin tolerance test. Early morning cut off cortisol levels for adrenal insufficiency in our center were validated for patients with hypothalamic-pituitary disorders as previously published [18]. Thyroid hormone deficiency was based on a low serum free thyroxine concentration (<11.0 pmol/l). Growth hormone deficiency was based on a low insulin-like growth factor 1 (IGF-1) Z-score (less than 2 SD) and/or an insufficient peak growth hormone (GH) concentration (<10 mU/l) in response to in-

sulin-induced hypoglycaemia or a peak growth hormone <18 mU/l during an arginine-GHRH test (<18 mU/l). Insufficiency of the pituitary– gonadal axis was defined in men as a testosterone concentration below 10 nmol/l, in premenopausal women (aged <50 years) as loss of menses and in postmenopausal women (aged >50 years) as LH and FSH concentrations below 15 mU/l. Diabetes insipidus was defined as the incapacity to properly concentrate urine (increased urine volume with low urine osmolality) in the face of a high plasma osmolality (and sodium). Biochemical control of adequacy of hormonal substitution treatment was judged by the physicians that were responsible for the care of the participating patients using free thyroxine, insulin-like growth factor 1 and testosterone levels where necessary. Other inclusion criteria were age 18 - 75 years, time interval of at least one year between study entry and tumor treatment with surgery and/or radiotherapy and adequate replacement of all other pituitary hormone deficiencies for at least six months prior to entry of the study.

Main exclusion criteria were documented severe cognitive impairment (MMSE < 24) [19], drug abuse or dependence, current psychiatric disorders, previous Cushing's disease, diabetes mellitus with medication known to be able to induce hypoglycemia (e.g. Sulfonylurea derivatives and insulin) and a history of frequent episodes of clinical hypocortisolism. Concomitant drugs known to interfere with HC metabolism, e.g. antiepileptics, were also not allowed [20].

All patients were tested in the period between May 2012 and January 2013. The baseline characteristics of the entire cohort of patients with SAI (n=135) did not differ from the presented study population, confirming the representativeness of our study population (data not shown).

Ethics Statement

The study protocol was approved by the local ethics committee at the UMCG, The Netherlands. Participants gave written informed consent before entering the study.

Cognitive tests

A battery of 11 standardized cognitive tests which resulted in 37 test scores covering 4 cognitive domains (memory, attention, executive functioning and social cognition) was used. This battery included: the Rivermead Behavioral Memory Test (RBMT), the 15 Words Test, Digit Span test, Rey Complex Figure test, the Test of Attentional Performance, verbal fluency tests, the Trail Making Test, and the Reading the Mind in the Eyes Test. See **Appendix** for a full description of tests and reference data.

Questionnaire

A common questionnaire on demographic and health-related data was used to assess educational level, social status, full-time/part-time employment, social security benefit and use of medication. Educational level was classified using a Dutch education system, comparable to the International Standard Classification of Education (ISCED) [21]. This scale ranges from 1 (elementary school not finished) to 7 (university level).

Procedure

The cognitive test battery took approximately 3 hours including a coffee break. All testing and

scoring of tests was performed by trained personnel under the supervision of two neuropsychologists (PB and JK).

Statistical analyses

All statistical analyses were performed using Statistical Package for the Social Sciences (SPSS, Inc., Armonk, NY, USA), version 20. Demographic data are presented as median, interquartile ranges (IQR), frequencies or percentages. Normality of data was analyzed using Q-Q plots. Since not all data were normally distributed, patients' cognitive performances were compared to the reference group by using non-parametric tests. Cognitive performance data were presented as mean *Z-score* with a non-parametric 95% confidence interval (CI) [22]. To analyze the influence of treatment characteristics, patients with at least one impaired score per cognitive domain (memory, attention, executive functioning and social cognition) were compared to patients with no impairments in the same domain in a univariate model. This model consisted of the variables: surgery, age at surgery, time since surgery, radiotherapy, age at radiotherapy, time since radiotherapy, dosis HC, mg HC/kg body weight and the use of thyroid hormone, growth hormone, sex hormone and desmopressin substitution. The use of sex hormones in men and women was not evaluated separately, since this would yield many different and small groups. A cognitive impairment on a test was defined as a performance equivalent to the performance of the lowest 10% of the reference samples [19]. Comparisons were only performed if the size of the groups was sufficient (i.e. $n > 5$). Categorical variables were analyzed using chi-square tests. Continuous variables were analyzed by the Mann-Whitney test. The two-tailed alpha level of <0.05 was considered statistically significant.

Results

Study population

Sixty patients with SAI participated in the present study (35 men and 25 women, mean (SD) age 52(14) years, range 19-73). Patients' characteristics are given in Table 1. Multiple pituitary insufficiencies were present in 57 patients (95%), while isolated ACTH deficiency was present in 3 patients (5%). Thirty one patients were treated for a pituitary adenoma (19 patients with a non-functioning macroadenoma, 6 patients with a macroprolactinoma, 6 patients with acromegaly). Five patients received treatment for a cyst with pituitary localization, 5 had a craniopharyngioma, 7 had a tumor distant from the pituitary gland (including germinomas, meningeomas, ENT tumor and a Primitive Neuroectodermal Tumor), and 5 patients had other acquired forms of ACTH deficiency (including Sheehan and traumatic brain injury). In addition, 7 had a congenital form of ACTH deficiency (6 combined, 1 isolated). Twenty-nine patients previously had undergone transsphenoidal surgery, 12 patients had undergone a craniotomy, 5 patients had primary radiotherapy and 21 patients had adjuvant radiotherapy. The median [IQR] cumulative daily HC dose was 25 [20; 30] mg. When corrected for kg body weight the median [IQR] HC dose was 0.31 [0.25; 0.35] mg. Most patients (70%) received twice daily dosing.

Cognitive tests

Table 2 summarizes mean *Z-scores* with a 95% CI for all cognitive measures. Tables 3a-d show the comparison of treatment characteristics between patients with or without an impairment in each cognitive domain separately.

Memory performance

Patients showed significantly lower scores on immediate, delayed and short-term verbal memory (RBMT and 15 words test). However, the mean delayed *Z-score* for the short story (RBMT) fell in the normal range when corrected for immediate memory. The performance on short-term memory (digit span forward) was significantly better compared to the reference sample. In addition, the non-verbal immediate and delayed memory scores of the Rey Complex Figure were significantly better compared to the reference sample. Of the entire patient sample, 80% (n=48) showed an impairment of at least one of the memory measures. Patients who received radiotherapy showed significantly more often a memory impairment than patients who received no radiotherapy (Table 3a).

Attention performance

Reaction times of patients in response to auditory stimuli of the divided attention task were significantly slower compared to reference data, whereas the reaction times of the visual stimuli fell in the normal range. During this task, patients made more omission errors compared to the reference sample. With regard to visual scanning, when patients had to press the key when a target stimuli was present, they reacted slower and had an increased variability in reaction times. When patients had to press the key when the target was not present, reaction times were faster although patients showed an increased variability of reaction time and also an increased number of commission errors. With regard to alertness, reaction times were significantly slower when patients had to respond as quickly as possible when a cross appeared (tonic alertness) and also when a warning tone preceded the appearance of the cross (phasic alertness). Of the entire sample, 85% (n=50) showed an impaired performance of at least one of the measures of attention. The time interval since surgery was significantly longer in impaired patients compared to unimpaired patients. Furthermore, patients in the impaired group used thyroid hormones more often compared to patients without an impaired performance on one of the measures of attention (Table 3b).

Executive functioning

Patients scored significantly worse on divergent thinking (fluency test) when they had to generate as many words as possible given a category (semantic fluency), but not when patients had to start with a specific letter (phonemic fluency). Assessment of psychomotor speed and cognitive flexibility (Trail Making Test, condition A and B) revealed that patients were significantly slower during both condition A and condition B. However, when the performance on condition B (cognitive flexibility) was corrected for the influence of condition A (psychomotor speed) the patient group and the reference sample did not differ anymore. Of the entire patient sample, 43% (n=26) showed an impairment on at least one of the measures of executive functioning (Table 3c).

Social cognition

Patients showed a significantly reduced performance on the recognition of emotions of the eye region as shown in pictures (the Reading the Mind in the Eyes Test) compared to the reference sample. In total 37% (n=22) showed an impairment in this domain. Time since radiotherapy was significantly longer in the impaired patient group compared to the unimpaired patients (Table 3d).

Table 1. Clinical characteristics of pituitary patients treated for adrenal insufficiency (N=60)

Basic characteristics	
Age (y), median [IQR]	55 [46; 62]
Sex (males/females)	35/25
Educational level (1/2/3/4/5/6/7) ♦	(0/2/1/9/28/18/2)
Age at diagnosis (y), median [IQR]	34 [21; 47]
Childhood onset/Adult onset of SAI	8/52
Surgery (n = 41)	
Transsphenoidal surgery/ Craniotomy (%)	71/29
Age at surgery (y), median [IQR]	40.8 [24.5; 50.5]
Average time since surgery (y), median [IQR]	11.4 [6.7; 21.0]
Patients with 2nd surgery (%)	13.3
Radiotherapy (n = 26)	
Pituitary radiotherapy/cranial irradiation/ radiotherapy for extracranial tumors (%)	77/19/4
Age at radiotherapy (y), median [IQR]	37.5 [23.6; 48.4]
Average time since radiotherapy (y), median [IQR]	14.6 [10.5; 24.8]
Hydrocortisone Treatment	
Total daily dose (mg/day), median [IQR]	25 [20; 30]
Dose/kg body weight (mg/kg), median [IQR]	0.31 [0.25; 0.35]
Number of daily dosings (1/2/3) (%)	5/70/25
Duration of hydrocortisone treatment (y), median [IQR]	11.4 [5.4; 21.9]
No. of hormonal replacements (1/2/3/4/5)	3/12/25/17/3
Thyroid hormone (% of patients substituted)	92
Growth hormone (% of patients substituted)	50
Growth hormone deficiency (% of patients unsubstituted)	33
Sex hormone (% of patients substituted)	55
Men: testosterone (% of patients substituted)	77
Premenopausal women, n=10: estrogens (% of patients substituted)	60
Postmenopausal women, n=15: estrogen	NA
Desmopressin (% of patients substituted)	22

♦ Educational level was classified using a Dutch education system, comparable to the International Standard Classification of Education (ISCED) [21]. This scale ranges from 1 (elementary school not finished) to 7 (university level).
IQR: Interquartile range, SAI: Secondary adrenal insufficiency, NA: Not applicable.

Table 2. Cognitive performance of pituitary patients treated for adrenal insufficiency (N=60)

	Mean *Z*-score [95%CI]	*P*-value*
Memory		
RBMT†		
Immediate memory	-0.79 [-1.05; -0.53]	<0.001
Delayed memory	-0.72 [-0.96; -0.48]	<0.001
Delayed corrected for immediate memory	-0.09 [-0.44; 0.26]	0.624
15 Words Test†		
Short-term memory	-0.31 [-0.57; -0.05]	0.017
Total immediate memory	-0.51 [-0.77; -0.25]	<0.001
Learning score	0.09 [-0.18; 0.36]	0.490
Delayed memory	-0.50 [-0.77; -0.23]	<0.001
Delayed corrected for total memory	-0.24 [-0.46; -0.02]	0.032
Recognition	-0.17 [-0.38; 0.04]	0.121
Digit Span forward†		
Short-term memory	0.27 [0.03; 0.51]	0.025
Rey Complex Figure ‡		
Immediate memory	0.66 [0.31; 1.02]	<0.001$^{\Delta}$
Delayed recall	0.60 [0.25; 0.95]	<0.001$^{\Delta}$
Attention		
Divided attention‡		
Reaction time auditory responses	-1.02 [-1.24; -0.80]	<0.001
Variability of reaction time auditory responses	0.09 [-0.18; 0.36]	0.490
Reaction time visual responses	-0.20 [-0.45; 0.05]	0.123
Variability of reaction time visual responses	-0.21 [-0.49; 0.07]	0.158
Number of omission errors	-0.51 [-0.75; -0.27]	<0.001
Number of commission errors	0.11 [-0.13; 0.35]	0.366
Visual scanning‡		
Reaction time for target stimuli	-0.74 [-0.97; -0.51]	<0.001
Variability of reaction time for target stimuli	-0.69 [-0.91; -0.46]	<0.001
Number of omission errors	-0.25 [-0.54; 0.04]	0.096
Reaction time for non-target stimuli	0.82 [0.59; 1.05]	<0.001
Variability of reaction time for non-target stimuli	-0.80 [-1.02; -0.58]	<0.001
Number of commission errors	-0.74 [-0.82; -0.66]	<0.001
Alertness‡		
Tonic alertness reaction time	-0.80 [-1.04; -0.56]	<0.001
Tonic alertness variablity in reaction time	-0.10 [-0.33; 0.13]	0.406
Phasic alertness reaction time	-1.07 [-1.25; -0.90]	<0.001
Phasic alertness variablity in reaction time	-0.16 [-0.38; 0.06]	0.163

	Mean *Z*-score [95%CI]	*P*-value*
Executive functioning		
Fluency test†		
Semantic fluency	-0.24 [-0.47; -0.01]	0.041
Phonemic fluency	-0.03 [-0.20; 0.14]	0.842
Digit Span backwards†		
Working memory	0.19 [-0.07; 0.45]	0.139
Trail Making Test‡		
Condition A, time	-0.64 [-0.92; -0.36]	<0.001
Condition A, no. of corrections (0/1)	55/5	
Condition B, time	-0.43 [-0.68; -0.18]	<0.001
Condition B, no. of corrections (0/1/2/3)	46/6/6/2	
Condition B/A	-0.12 [-0.36; 0.12]	0.327
Social cognition †‡		
Reading the Mind in the Eyes Test	-0.94 [-1.21; -0.67]	<0.001

Comparing patients' performance to reference data; †: verbal test; ‡: non-verbal test; CI: Confidence Interval; RBMT: Rivermead Behavioral Memory Test; $^{\Delta}$Scores for n = 56 patients (4 patients were excluded from analysis because of an impaired copy drawing, <10th percentile) (See Appendix)

Table 3a. Comparison of treatment characteristics between patients with or without memory impairment

	Impaired patients		Unimpaired patients		*P*-value
	N	Median [IQR]/frequencies	N	Median [IQR]/frequencies	
Surgery (yes/no)	48	34/14	12	7/5	0.405
Age at surgery (y)	34	41.6 [26.3;50.5]	7	38.1 [20.1;50.7]	0.703
Time since surgery (y)	34	11.9 [6.2;22.2]	7	10.7 [10.6;17.0]	0.862
Radiotherapy (yes/no)	48	25/23	12	1/11	**0.006**
Dosis hydrocortisone (mg/day)	48	30 [20;30]	12	20 [20;30]	0.328
Mg hydrocortisone/kg body weight	48	0.31 [0.26;0.35]	12	0.30 [0.23;0.38]	0.919
Thyroid hormone substitution (yes/no)	48	44/4	12	11/1	1.000
Growth hormone substitution (yes/no)	48	24/24	12	6/6	1.000
Sex hormone substitution (yes/no)	48	28/20	12	5/7	0.299
Desmopressin substitution (yes/no)	48	8/40	12	5/7	0.060

IQR: Interquartile range; Age at radiotherapy and time since radiotherapy were not shown, since the number of patients in the unimpaired group was insufficient (i.e. <5)

Table 3b. Comparison of treatment characteristics between patients with or without attention impairment

	Impaired patients		Unimpaired patients		*P*-value
	N	Median [IQR]/frequencies	N	Median [IQR]/frequencies	
Surgery (yes/no)	50	34/16	9	7/2	0.558
Age at surgery (y)	34	38.1 [20.7;51.6]	7	42.9 [31.6;49.6]	0.729
Time since surgery (y)	34	12.9 [7.4;25.7]	7	5.8 [2.9;10.7]	**0.024**
Radiotherapy (yes/no)	50	23/27	9	3/6	0.418
Dosis hydrocortisone (mg/day)	50	25 [20;30]	9	30 [20;30]	0.964
Mg hydrocortisone/kg body weight	50	0.31 [0.27;0.35]	9	0.28 [0.23;0.38]	0.800
Thyroid hormone substitution (yes/no)	50	48/2	9	7/2	**0.045**
Growth hormone substitution (yes/no)	50	27/23	9	3/6	0.254
Sex hormone substitution (yes/no)	50	27/23	9	6/3	0.418
Desmopressin substitution (yes/no)	50	11/39	9	2/7	0.988

IQR: Interquartile range; Age at radiotherapy and time since radiotherapy were not shown, since the number of patients in the unimpaired group was insufficient (i.e. <5) ; Note: Data are derived from 59 patients (see Appendix).

Table 3c. Comparison of treatment characteristics between patients with or without executive functioning impairment

	Impaired patients		Unimpaired patients		*P*-value
	N	Median [IQR]/frequencies	N	Median [IQR]/frequencies	
Surgery (yes/no)	26	18/8	34	23/11	0.896
Age at surgery (y)	18	36.3 [18.9; 50.5]	23	42.9 [28.5;50.7]	0.415
Time since surgery (y)	18	16.0 [6.0;23.0]	23	10.7 [6.3;17.0]	0.462
Radiotherapy (yes/no)	26	12/14	34	14/20	0.700
Age at radiotherapy (y)	12	28.0 [16.5;45.8]	14	42.0 [30.1;55.1]	0.064
Time since radiotherapy (y)	12	19.5 [9.0;31.4]	14	12.0 [9.9;19.9]	0.280
Dosis hydrocortisone (mg/day)	26	27.5 [20;30]	34	25 [20;30]	0.797
Mg hydrocortisone/kg body weight	26	0.31 [0.27;0.35]	34	0.29 [0.24;0.35]	0.328
Thyroid hormone substitution (yes/no)	26	25/1	34	30/4	0.271
Growth hormone substitution (yes/no)	26	13/13	34	17/17	0.100
Sex hormone substitution (yes/no)	26	16/10	34	17/17	0.373
Desmopressin substitution (yes/no)	26	5/21	34	8/26	0.689

IQR: Interquartile range; Note: Condition A of the Trail Making Test is a measure of psychomotor speed and therefore not included in this analyses.

Table 3d. Comparison of treatment characteristics between patients with or without social cognition impairment

	Impaired patients		Unimpaired patients		*P*-value
	N	Median [IQR]/frequencies	N	Median [IQR]/frequencies	
Surgery (yes/no)	22	14/8	38	27/11	0.552
Age at surgery (y)	14	36.3 [20.7;52.0]	27	42.3 [28.1;50.3]	0.700
Time since surgery (y)	14	18.0 [10.1;29.9]	27	10.6 [4.2;17.0]	0.070
Radiotherapy(yes/no)	22	10/12	38	16/22	0.651
Age at radiotherapy (y)	10	32.2 [20.8;44.0]	16	42.0 [28.8;53.2]	0.140
Time since radiotherapy (y)	10	20.7 [13.1;34.8]	16	11.6 [6.1;17.9]	**0.035**
Dosis hydrocortisone (mg/day)	22	30 [20;30]	38	25 [20;30]	0.432
Mg hydrocortisone/kg body weight	22	0.31 [0.27;0.36]	38	0.31 [0.25;0.35]	0.466
Thyroid hormone substitution (yes/no)	22	22/0	38	33/5	0.076
Growth hormone substitution (yes/no)	22	12/10	38	18/20	0.592
Sex hormone substitution (yes/no)	22	14/8	38	19/19	0.306
Desmopressin substitution (yes/no)	22	4/18	38	9/29	0.618

IQR: Interquartile range

Discussion

This study demonstrated that patients treated for SAI showed impairments in the cognitive domains of memory, attention, executive functions and social cognition compared to reference data of the general population. There is, however, a pattern of selective deficits that can be detected with some cognitive functions being impaired while others remain preserved. Furthermore, we found that various forms of treatment in this patient population related to impairments in different cognitive domains.

From previous studies in healthy volunteers, it is known that GC can impair aspects of cognition that depend upon the integrity of the hippocampus, i.e. declarative memory (memory we have conscious access to) [23]. These results are confirmed by the present study which shows that 80% of SAI patients display an impairment on at least one memory test. The present study goes, however, beyond previous findings and demonstrates a discrepancy between verbal and non-verbal declarative memory in patients with SAI. With regard to verbal memory it was found that in particular the encoding and retrieval of information is impaired, with the impairment of retrieval being specific for incoherent information compared to coherent information. The recognition of verbal information appears to be unimpaired, most likely because this task requires only the relatively simple decision as to whether specific information was encountered before or not. The finding of an intact recognition while the encoding and retrieval of verbal information is affected, is in line with a study from de Quervain and colleagues [4].

The patients with SAI in this study showed no impairments on the non-verbal memory tasks and performed even significantly better than the reference samples. One explanation for this might be that exogenous cortisol leads to more positive-going waveforms over the right than the left hemisphere, which are involved in non-verbal and verbal memory, respectively [11]. Furthermore, if verbal memory is more affected by exogenous cortisol than non-verbal memory, patients might have learned to cope with this by applying more visual compensation techniques. Another explanation for the better performance of the patient group in the non-verbal memory test compared to the reference sample could be that the patient group put more effort in this test compared to the reference group [24]. No general memory deficit was thus found in the present study, indicating selective impairments which might at least in part be determined by the material that has to be processed (verbal versus non-verbal and coherent versus incoherent).

In the cognitive domain of attention, 85% of SAI patients showed an impairment. Patients displayed in particular a slower processing speed or increased reaction times on most tests. A discrepancy was, however, found for the divided attention task during which both auditory and visual stimuli need to be processed simultaneously. Patients showed significantly slower reaction times in response to the auditory stimuli but not in response to visual stimuli. Apparently, patients automatically pay more attention to visual stimuli compared to auditory stimuli, which is in accordance with the discrepancy observed between verbal and non-verbal (i.e. visual) memory. As suggested earlier, patients might have learned visual compensation techniques and unconsciously pay more attention to visual stimuli. Furthermore, it was found that patients do not pay sufficient attention to relevant stimuli, i.e. they display a significantly higher number of omission errors (lack of response to target stimuli) during the divided attention task. In the selective atten-

tion task, patients showed more commission errors (responses to non-target stimuli during the visual scanning task) which could be indicative of an increased impulsivity. Taken together, patients with SAI have a slower processing speed in addition to a potentially increased distractibility and impulsivity.

With regard to executive functioning, selective deficits were also found on semantic fluency whereas phonemic fluency was unimpaired. This pattern was previously detected in patients with Parkinson's disease [25,26]. It has been suggested that even though both fluency measures depend on the integrity of executive functions, semantic fluency tests are also influenced by the integrity of semantic memory [25]. The difference between semantic and phonemic fluency might therefore be (partly) explained by impairments in declarative memory which were also found in the present study. Other aspects of executive functioning, i.e. cognitive flexibility and working memory were not impaired compared to the reference data of the general population. However, when a more clinical individual approach was used the results show that 43% of patients treated for SAI have at least one impairment in the domain of executive functioning. Impairments in executive functioning have previously been demonstrated in healthy participants receiving different doses of HC by infusion [12]. The apparent discrepancy between no major average group effect on the one hand and the higher prevalence of at least one impairment for an individual patient on the other hand justifies a more personal approach for cognitive evaluation.

Impaired performances were also found in the domain of social cognition. Social cognition refers to the mental operations that underlie social interactions, including the perception of emotions, interpreting social behavior, and generating responses to the intentions, dispositions, and behaviors of others [27]. An impaired performance in this domain was already found in women, with higher levels of cortisol leading to decreased performances on social cognition tasks [28]. The current study found a decreased social cognitive performance in both men and women with SAI. Furthermore, when a more clinical approach was used, 37% of pituitary patients treated for adrenal insufficiency showed an impairment. Social cognition is, however, a complex cognitive function which was not examined in detail in the present study. Future studies should therefore put a bigger emphasis on the domain of social cognition in patients with SAI.

Different treatment modalities and treatment related factors were associated to cognition in patients with SAI. In exploratory analyses, the use of thyroid hormone was more frequent in patients with impaired attention, whereas the use of desmopressin was borderline significantly associated with better memory performance. The association between thyroid hormone and cognition is in line with a recently published study by Beydoun *et al.* [29]. The latter is consistent with the study of Vawter *et al.* who also found that a vasopressin fragment improved long-term and short-term memory, although this was found in rats [30]. In addition, glucocorticoid dose corrected for body weight was higher in patients with impaired psychomotor speed (TMT-A, data not shown), a finding that warrants further investigation. Also, ten patients without growth hormone substitution (33%) had IGF-1 *Z-scores* < -2, indicating unsubstituted severe GH deficiency in this population. This is usually the consequence of an absence of GH deficiency related symptoms or lack of patient benefit on GH treatment. Untreated GH deficiency might also have influenced results [31-33]. However, except for one outcome measure, the patients with IGF-1 *Z-scores* <-2 did not perform significantly different compared to patients with IGF-1 *Z-scores* within the normal range (i.e. patients with untreated GH deficiency scored significantly lower on the semantic fluency test,

data not shown). Both radiotherapy and surgery were found to be associated with poorer performance on memory, executive functioning, attention and social cognition. In our previous studies [34,35,] we did not reveal such effects of radiotherapy and surgery when comparing the group of pituitary patients to reference data of the general population, also not on self reported cognition [36]. This discrepancy might be explained by the different tests used to examine cognition in these studies. One could argue that a Bonferroni correction for multiple testing should have been performed. However, although we performed multiple comparisons, the cognitive domains assessed in the current study are not fully dependent on each other. Furthermore, this was a hypothesis generating analysis and not a hypothesis affirmative research. Therefore, we show absolute *p values* and demonstrate that different cognitive domains relate to different treatment characteristics. The implication of these findings is that certain treatments do not globally affect cognition but may rather result in selective impairments. This is a conclusion that does not depend on *p values* with a certain statistical cut-off , but rather on different overall patterns.

To our knowledge, this is the first study on the effects of long-term physiological doses of HC in patients with SAI in relation to cognition as examined with a comprehensive cognitive battery consisting of psychometric tests. The results seem to be consistent with earlier findings indicating an impaired cognitive performance in healthy subjects with altered glucocorticoid levels [4-7, 9, 10, 12, 13]. Some study limitations, however, need to be addressed. First, this is a cross-sectional study. Cross sectional studies are effective when evaluating a large group of patients and the associations with treatment characteristics. However, we cannot draw any conclusions about HC exposure and cognitive outcome, since exposure and outcome are determined at the same time. Secondly, a healthy control group was not included in the current study. Therefore it is not clear to what extent individuals in the general population are impaired on at least one test in the specific cognitive domains. However, the magnitude of differences in *Z-scores* as shown in Table 2 highly suggest that impairments on at least one test in a specific domain is not as prevalent as in our patient population. Future studies on cognition should include a control group, which consists of healthy individuals who are matched for age, sex and education to the group of patients. Thirdly, we have to bear in mind that the average daily dose of HC based on body weight of the patient group included in the present study is somewhat higher than is generally accepted as desirable [37]. However, as experienced in our own outpatient group, it can be difficult to change treatment strategies because patients are used to their treatment regimes for many years and other treatment regimens might cause undesirable symptoms (e.g. sleep disturbances and headache). Lastly, in this study patients with different surgery and radiotherapy techniques were examined. Although the aim of the study was to examine cognitive functioning in a representative group of patients with SAI as they are treated nowadays (i.e. with different treatment characteristics), one should be aware of the fact that different surgery and radiotherapy techniques might have different effects on cognition, potentially obscuring subtle effects.

In conclusion, no global cognitive impairments were found in pituitary patients treated for adrenal insufficiency, but rather various selective deficits, in particular on verbal cognitive tasks, with some cognitive functions being impaired while others remaining preserved. The results of the present study are of importance for patient management, since cognitive impairments frequently affect every day functioning.

References

1 Hahner S, Loeffler M, Fassnacht M, Weismann D, Koschker AC, et al. 2007 Impaired subjective health status in 256 patients with adrenal insufficiency on standard therapy based on cross-sectional analysis. *J Clin Endocrinol Metab* 92(10): 3912-3922.

2 Bergthorsdottir R, Leonsson-Zachrisson M, Oden A, Johannsson G 2006 Premature mortality in patients with Addison's disease: A population-based study. *J Clin Endocrinol Metab* 91(12): 4849-4853.

3 Gallagher P, Reid KS, Ferrier IN 2009 Neuropsychological functioning in health and mood disorder: Modulation by glucocorticoids and their receptors. *Psychoneuroendocrinology* 34 Suppl 1: S196-207.

4 de Quervain DJ, Roozendaal B, Nitsch RM, McGaugh JL, Hock C 2000 Acute cortisone administration impairs retrieval of long-term declarative memory in humans. *Nat Neurosci* 3(4): 313-314.

5 Kirschbaum C, Wolf OT, May M, Wippich W, Hellhammer DH 1996 Stress- and treatment-induced elevations of cortisol levels associated with impaired declarative memory in healthy adults. *Life Sci* 58(17): 1475-1483.

6 Lupien SJ, Gaudreau S, Tchiteya BM, Maheu F, Sharma S, et al. 1997 Stress-induced declarative memory impairment in healthy elderly subjects: Relationship to cortisol reactivity. *J Clin Endocrinol Metab* 82(7): 2070-2075.

7 Lupien SJ, Wilkinson CW, Briere S, Ng Ying Kin NM, Meaney MJ, et al. 2002 Acute modulation of aged human memory by pharmacological manipulation of glucocorticoids. *J Clin Endocrinol Metab* 87(8): 3798-3807.

8 Lupien SJ, Fiocco A, Wan N, Maheu F, Lord C, et al. 2005 Stress hormones and human memory function across the lifespan. *Psychoneuroendocrinology* 30(3):225-242.

9 Newcomer JW, Craft S, Hershey T, Askins K, Bardgett ME 1994 Glucocorticoid-induced impairment in declarative memory performance in adult humans. *J Neurosci* 14(4):2047-2053.

10 Newcomer JW, Selke G, Melson AK, Hershey T, Craft S, et al. 1999 Decreased memory performance in healthy humans induced by stress-level cortisol treatment. *Arch Gen Psychiatry* 56(6): 527-533.

11 Alhaj HA, Massey AE, McAllister-Williams RH 2008 Effects of cortisol on the laterality of the neural correlates of episodic memory. *J Psychiatr Res* 42(12): 971-981.

12 Lupien SJ, Gillin CJ, Hauger RL 1999 Working memory is more sensitive than declarative memory to the acute effects of corticosteroids: A dose-response study in humans. *Behav Neurosci* 113(3): 420-430.

13 McCormick CM, Lewis E, Somley B, Kahan TA 2007 Individual differences in cortisol levels and performance on a test of executive function in men and women. *Physiol Behav* 91(1):87-94.

14 Joels M 2008 Functional actions of corticosteroids in the hippocampus. *Eur J Pharmacol* 583(2-3): 312-321.

15 Sanchez MM, Young LJ, Plotsky PM, Insel TR 2000 Distribution of corticosteroid receptors in the rhesus brain: Relative absence of glucocorticoid receptors in the hippocampal formation. *J Neurosci* 20(12): 4657-4668.

16 van Overwalle F 2009 Social cognition and the brain: A meta-analysis. *Hum Brain Mapp* 30(3): 829-858.

17 Li C, Chen K, Han H, Chui D, Wu J 2012 An FMRI study of the neural systems involved in visually cued auditory top-down spatial and temporal attention. *PLoS One* 7(11): e49948.

18 Dullaart RP, Pasterkamp SH, Beentjes JA, Sluiter WJ 1999 Evaluation of adrenal function in patients with hypothalamic and pituitary disorders: Comparison of serum cortisol, urinary free cortisol and the human-corticotrophin releasing hormone test with the insulin tolerance test. *Clin Endocrinol (Oxf)* 50(4): 465-471.

19 Lezak, MD, Howieson D, Loring DW 2004 Neuropsychological Assessment, 4 ed. New York: Oxford University Press.

20 Debono M, Price JN, Ross RJ 2009 Novel strategies for hydrocortisone replacement. *Best Pract Res Clin Endocrinol Metab* 23(2): 221-232.

21 United Nations Educational. Scientific and Cultural Organization. International Standard Classifications of Education ISCED. 2006.

22 Campbell MJ, Gardner, MJ 1988 Calculating confidence intervals for some non-parametric analysis. *British Medical Journal*; 196: 1454-1456.

23 Jameison K, Dinan TG 2001 Glucocorticoids and cognitive function: From physiology to pathophysiology. *Hum Psychopharmacol* 16(4): 293-302.

24 Reedy SD, Boone KB, Cottingham ME, Glaser DF, Lu PH, et al. 2013 Cross validation of the lu and colleagues 2003 Rey-Osterrieth Complex Figure Test effort equation in a large known-group sample. *Arch Clin Neuropsychol* 28(1): 30-37.

25 Henry JD, Crawford JR 2004 Verbal fluency deficits in Parkinson's disease: A meta-analysis. *J Int Neuropsychol Soc* 10(4): 608-622.

26 Koerts J, Meijer HA, Colman KS, Tucha L, Lange KW, et al. 2013 What is measured with verbal fluency tests in Parkinson's disease patients at different stages of the disease? *J Neural Transm* 120(3): 403-411.

27 Davis MC, Lee J, Horan WP, Clarke AD, McGee MR, et al. 2013 Effects of single dose intranasal oxytocin on social cognition in schizophrenia. *Schizophr Res* 147(2-3): 393-7.

28 Smeets T, Dziobek I, Wolf OT 2009 Social cognition under stress: Differential effects of stress-induced cortisol elevations in healthy young men and women. *Horm Behav* 55(4): 507-513.

29 Beydoun MA, Beydoun HA, Kitner-Triolo MH, Kaufman JS, Evans MK, *et al.* 2013 Thyroid hormones are associated with cognitive function: Moderation by sex, race and depressive symptoms. *J Clin Endocrinol Metab* 98(8): 3470-81.

30 Vawter MP, De Wied D, Van Ree JM 1997 Vasopressin fragment, AVP-(4-8), improves long-term and short-term memory in the hole board search task. *Neuropeptides* 31(5): 489-494.

31 Sathiavageeswaran M, Burman P, Lawrence D, Harris AG, Falleti MG, et al. 2007 Effects of GH on cognitive function in elderly patients with adult-onset GH deficiency: A placebo-controlled 12-month study. *Eur J Endocrinol* 156(4): 439-447.

32 Oertel H, Schneider HJ, Stalla GK, Holsboer F, Zihl J 2004 The effect of growth hormone substitution on cognitive performance in adult patients with hypopituitarism. *Psychoneuroendocrinology* 29(7): 839-850.

33 Falleti MG, Maruff P, Burman P, Harris A 2006 The effects of growth hormone (GH) deficiency and GH replacement on cognitive performance in adults: A meta-analysis of the current literature. *Psychoneuroendocrinology* 31(6): 681-691.

34 Brummelman P, Elderson MF, Dullaart RP, van den Bergh AC, Timmer CA, et al. 2011 Cognitive functioning in patients treated for nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Clin Endocrinol (Oxf)* 74(4): 481-487

35 Brummelman P, Sattler MG, Meiners LC, Elderson MF, Dullaart RP, et al. 2012 Cognitive performance after postoperative pituitary radiotherapy: A dosimetric study of the hippocampus and the prefrontal cortex. *Eur J Endocrinol* 166(2): 171-179

36 van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, et al. 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated with nonfunctioning pituitary adenoma. *Int J Radiat Oncol Biol Phys* 68(4): 986-991

37 Mah PM, Jenkins RC, Rostami-Hodjegan A, Newell-Price J, Doane A, et al. 2004 Weight-related dosing, timing and monitoring hydrocortisone replacement therapy in patients with adrenal insufficiency. *Clin Endocrinol (Oxf)* 61(3): 367-75.

Appendix. Full description of cognitive tests and reference data

The following cognitive domains were assessed using several standardized and validated cognitive tests: memory, attention, executive functioning and social cognition.

Memory

During the Rivermead Behavioral Memory Test (RBMT) [1], a short story was read out loud to patients who were then asked to immediately recall as many details as possible from the story. This allowed the calculation of an immediate memory score for coherent verbal information. A delayed memory score for coherent verbal memory was determined based on the number of details of the story recalled by patients after a period of about 30 minutes. Verbal memory was also assessed with the 15 Words Test, which is a Dutch equivalent of the Rey Auditory Verbal Learning Test [2]. During this test, 15 words were presented five times. After each trial, patients were asked to name immediately the words they remembered. This allowed the calculation of three different scores describing immediate memory. The *short-term memory score* is based on the number of words patients were able to name after the first presentation of the word list. The *total immediate memory score* represents the total number of words the patients remembered over the five trials. The *learning score* describes the difference between the number of words remembered in the third trial in comparison with the first trial. Besides immediate memory, delayed memory was measured with the 15 Words Test. The *delayed memory score* is based on the number of words a patient could recall after a period of about 30 minutes. To correct for the total immediate memory, there was also a *delayed corrected for total memory score*. The last part of the test was a recognition task. A total of 30 words were presented to the patient, of which 15 words were presented during the previous five trials and of which 15 words were new to the patient. The patients had to decide whether the words were new or familiar, which allowed the calculation of a *recognition memory score*.

To assess short-term memory, the Digit Span Forward test was used [3]. Patients were presented with a sequence of digits and immediately had to repeat the digits in the same order as they were presented. The test started with sequences of two digits which expanded until the maximum of nine digits. When a patient could not correctly repeat two sequences of the same length, the test was aborted. The total number of correctly repeated sequences was registered. Visual memory was assessed with the Rey Complex Figure Test [4]. During this test, a complex geometrical figure was placed in front of patients, who were instructed to copy the figure as accurately as possible. After a 3-minute delay, patients were asked to reproduce the figure from memory. Approximately 15 minutes later, patients were again asked to reproduce the figure from memory. The number of details correctly recalled after 3 and 15 minutes were used to calculate an immediate and delayed memory score. If patients had an impaired copy drawing (<10th percentile) measures of immediate and delayed memory were excluded for analysis. An impaired copy drawing might indicate problems with visuoconstructive functions causing unreliable scores for immediate and delayed memory.

Attention

The Test of Attentional Performance is a computerized test consisting of several subtests [5]. In the present study, the subtests divided attention, visual scanning and alertness were used. Test instructions were presented on a computer screen and read out loud by the test examiner. In order to familiarize the participants with the tasks, a brief sequence of practice trials preceded each test.

Divided attention is needed when two (or more) tasks have to be performed simultaneously. In the visual task, a series of matrices was presented in the center of the computer screen. Each matrix (4x4) consisted of a regular array of sixteen dots and crosses. When four of these crosses formed a square, patients had to press a key as quickly as possible. The acoustic task consisted of a high (2000 Hz) and low (1000 Hz) tone which were presented alternately according to the synchronous rhythm of the changing positions of the crosses. When the same tone occurred twice in a row, patients had to press a key as quickly as possible. A total of 100 visual and 200 acoustic stimuli was presented including 17 visual and 16 acoustic targets. Reaction time of correct responses, variability in reaction time and the number of omission (lack of response to target stimuli) and commission (responses to non-target stimuli) errors were calculated and represented measures of divided attention.

During the visual scanning task a series of 5 by 5 matrices were presented. Each matrix consisted of a regular array of 25 squares which each had an opening on one side (top, bottom, left or right side). A square with an opening on the top was defined as the target stimulus. The target stimulus occurred in 50% of matrices and was randomly distributed across the matrix. Patients were asked to press the left response button as quickly as possible when a matrix contained the target stimulus and to press the right response button if the target stimulus was not present. A total of 50 trials was presented (25 target trials and 25 non-target trials). Reaction times of correct responses, variability of reaction time and the number of commission and omission errors were calculated and represented measures of selective attention, i.e. attention for the target stimulus and not for non-target stimulus.

During the alertness task, reaction times were measured during two conditions. The first condition concerned a simple reaction time measurement, during which a cross appeared on the screen at random varying intervals. Patients were instructed to respond as quickly as possible by pressing a key when the cross appeared (tonic alertness task). In the second condition, a warning tone preceded the appearance of the stimulus (phasic alertness task). A total of 40 trials were presented, 20 trials with warning tone and 20 trials without warning tone. The time span between the warning tone and the appearance of the stimulus was random (between 300 and 700 ms). Measures of tonic and phasic alertness were calculated on the basis of reaction times of patients. In addition, the variability of reaction time and the number of omission errors were measured.

In this study, data of the Test of Attentional Performance were derived from 59 patients due to a technical problem with the computer during one patient visit.

Executive functioning

Divergent thinking is the ability to approach a task or situation in different ways. So-called fluency tests rely on divergent thinking and evaluate the spontaneous production of words under

restricted search conditions [6]. During the first fluency test applied in the present study, the semantic subtest, patients were asked to name as many animals as possible within one minute. Participants were not allowed to name the same word twice. The total number of correctly produced animals was counted. In a second fluency test, the phonemic subtest, patients were asked to name as many words as possible in one minute, starting with a specific letter. All existing nouns were allowed. However, personal names and names of places were not allowed. This part was administered three times, each time using a different letter, namely D, A and T [7]. For both subtests, the total number of correctly produced words was counted. To assess working memory, the Digit Span Backward test was used [3]. Patients were presented with a sequences of digits, which they had to repeat backwards. The test started with sequences of two digits which expanded until the maximum of eight sequences. When a patient failed to correctly repeat two sequences of the same length, the test was aborted. The total number of sequences that was repeated correctly was registered.

The Trail Making Test assesses visual scanning, speed of processing (condition A), cognitive flexibility (condition B) [8]. During condition A, patients were asked to connect 25 circles containing numbers as quickly as possible in ascending order. Part B consisted of 25 circles, containing both numbers and letters. Patients were again required to connect the circles as fast as possible in ascending order, this time alternating between numbers and letters i.e. 1-A-2-B-3-C, etc.. The target measure for cognitive flexibility was the performance on condition B corrected for condition A (Trail Making Test B/A).

Social cognition

Social cognition can be defined as the mental operations that underlie social interactions, including perceiving, interpreting, and generating responses to the intentions, dispositions, and behaviors of others [9]. In this study, social cognition was assessed with the Reading the Mind in the Eyes Test (RMET) [10]. During this test, patients were presented with 36 photographs, one after each other, of the eye-region of the face. Along with each pair of eyes, four words describing an emotion were presented. Patients were asked to choose which word described best what the person in the picture is feeling or thinking. To exclude any influence of difficulties with word comprehension, a glossary with the presented words was made available.

Reference data: Healthy control subjects

The performances of patients were compared to published normative datasets from healthy populations.

Memory

The results of the RBMT were compared to a healthy reference group (n=344) consisting of participants aged 17-89 [11]. The scores of patients were corrected for age, sex and education. In the study by Schmand *et al.*, participants were presented with two stories of comparable length. However, in the present study, patients were presented with only one story. To compensate for this difference, the score obtained in the present study was doubled. Reference data for the 15 Words Test were derived from control subjects (n=847) consisting of participants aged 14-87 [11] for the

measures total immediate memory, delayed memory and delayed corrected for total memory. Reference data for the measures short-term memory, learning score and recognition of the 15 Words Test were derived from control subjects of the Maastricht Aging Study [2]. The scores were corrected for age, sex and education. The results of the Digit Span test were compared to the reference group as described in the Wechsler Memory Scale Manual (n=316). These scores were corrected for age [3]. Patients' scores on the Rey Complex Figure were compared to reference data as provided in the manual of the Rey Complex Figure Test and Recognition Trial [12] and were also corrected for age.

Attention

Norm data for the divided attention, visual scanning and alertness subtests were provided by the test authors within the Test of Attentional Performance Version 2.2. [5]. A correction for age and sex was applied.

Executive functioning

The results of the semantic subtest of the Verbal Fluency Test were compared to a reference group (n=464) consisting of healthy participants aged 17-90 [11]. The scores were corrected for age and education. The scores of the phonemic subtest of the Verbal Fluency Test were compared to a reference group (n=570) consisting of participants aged 17-90 [11] and corrected for education. The results of the TMT were compared to a reference group (n=478), consisting of healthy participants aged 17-90 [11]. The scores were corrected for age, sex and education.

Social cognition

The performance on the RMET was compared to a healthy reference group (n=200) [13] and corrected for sex.

References

1 Wilson BA, Cockburn J, Baddeley AD 1985 The Rivermead Behavioural Memory Test. Bury St Edmunds: Thames Valley Test Company.

2 Van der Elst W, van Boxtel MP, van Breukelen GJ, Jolles J 2005 Rey's verbal learning test: Normative data for 1855 healthy participants aged 24-81 years and the influence of age, sex, education, and mode of presentation. *J Int Neuropsychol Soc* 11(3): 290-302.

3 Wechsler D 1987 Wechsler Memory Scale Revised. The Psychological Corporation.

4 Rey A 1941 L'examen psychologique dans les cas d'encéphalopathie traumatique. *Archives de Psychologie*, 28, 286-340.

5 Zimmermann P, Fimm B 2007 Test of Attentional Performance Version 2.2. Psychologische Testsysteme.

6 Straus E, Sherman E, Spreen O 2006 A compendium of neuropsychological test: administration norms and commentary. New York, USA: University Oxford Press.

7 Schmand B, Groenink SC, van den Dungen M 2008 Letter fluency: Psychometric properties and dutch normative data. *Tijdschr Gerontol Geriatr* 39(2): 64-76.

8 Reitan RM 1958 Validity of the Trail Making Test as an indicator of organic brain damage. *Percept Mot Skills*, 8, 271-276.

9 Davis MC, Lee J, Horan WP, Clarke AD, McGee MR, et al. 2013 Effects of single dose intranasal oxytocin on social cognition in schizophrenia. *Schizophr Res*: 147(2-3): 393-7.

10 Baron-Cohen S, Wheelwright S, Hill J, Raste Y, Plumb I 2001 The "reading the mind in the eyes" test revised version: A study with normal adults, and adults with asperger syndrome or high-functioning autism. *J Child Psychol Psychiatry* 42(2): 241-251.

11 Schmand B, Houx P, de Koning I 2012 Normen neuropsychologische tests 2012. Nederlands Instituut van Psychologen.

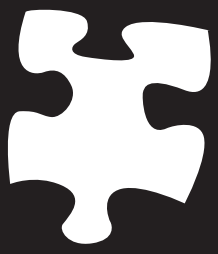
12 Meyers JE, Meyers KR 1995. RCFT Rey Complex Figure Test and Recognition Trial: Professional Manual. Ltz, FL. Psychological Assessment Recources..

13 Vellante M, Baron-Cohen S, Melis M, Marrone M, Petretto DR, Masala C, Preti A 2013 The "Reading the Mind in the Eyes" test: systematic review of psychometric properties and a validation study in Italy. *Cognitive Neuropsychiatry*, 18(4):326-354.

Discussion and Summary

Chapter 7

General discussion and summary



General discussion and conclusions

In the present thesis, we focused on the effects of different treatment options on cognition in patients with pituitary diseases. Reliable information on the possible side effects of different treatment options is of particular importance for clinicians and patients to allow an informed discussion and decision on treatment strategies. Furthermore, knowledge of the impact of specific treatments on cognition might also serve as a basis for theory driven approach to the evaluation of treatment strategies.

The effects of radiotherapy on cognition in patients with a nonfunctioning pituitary adenoma

The aim of **chapter 2** was to assess memory and executive functioning in a large (n = 84) homogeneous group of NFA patients. We specifically focused on NFA patients, since inconsistent results of previous studies were most likely the consequence of heterogeneous and small patient groups. In subjects with hormone-producing tumors, like Cushing's disease, the hormone overproduction itself may also have an effect on cognition. The second aim of this chapter was to compare patients who received both surgery and radiotherapy with those who only underwent surgery. It was found that NFA patients displayed a worse cognitive performance compared to reference populations on both verbal memory and executive functioning. However, the effects of radiotherapy were at most very small and unlikely to be of clinical relevance.

Since relatively small effects of radiotherapy on cognition in NFA patients could not be excluded (e.g. due to different radiotherapy techniques and therefore dose distribution), we refined the strategy and related radiation exposure of brain areas of interest (i.e. the hippocampus and the prefrontal cortex) to cognitive performance typically associated with these brain areas **(chapter 3)**. This follow-up study showed that current multiple field radiotherapy techniques do not have a significant negative effect on memory and executive functioning in patients with NFA. Also, a dose-response relationship could not be established. Although this was found in a group of patients with similar dose prescriptions, it can be argued that the different multiple field radiotherapy techniques applied seem to operate within safe radiotherapy dose boundaries.

White matter lesions and cerebral atrophy are well known radiologically detectable brain abnormalities following radiotherapy, which are reported to be associated with diminished cognitive performance [1,2]. So far, imaging studies on the association between brain abnormalities after radiotherapy and cognition have focused on patients without fixed tumor localization. These studies particularly included patients with low grade gliomas [1-3] in whom different cognitive domains are affected because of varying tumor localization, which leads to heterogeneous results. Therefore, we assessed and compared brain abnormalities on imaging in relation to cognitive impairments in NFA patients who have a fixed tumor location **(chapter 4)**. The results of this study showed that the frequency of brain abnormalities did not differ between NFA patients with and without cognitive impairments in the domains of verbal memory and executive functioning.

In conclusion, the results of chapters 2, 3 and 4 indicate that current multiple field radiotherapy techniques and fractionated radiation dose regimens – applied in addition to surgery - do not have a major negative effect on memory and executive functioning in patients with NFA.

A possible explanation for our findings might be that there is some kind of dose related threshold for radiotherapy to injure the brain and cause cognitive impairment, suggesting that only above this threshold a dose-response relationship can be found. The results of chapters 2, 3 and 4 indicate that all radiotherapy techniques which have been used over the years for the treatment of NFA patients in our center appear to operate within safe radiotherapy dose boundaries and do not have a major effect on memory and executive functioning.

Another explanation could also be that the adverse effects of radiotherapy have not become evident yet, since deterioration in cognitive functions might appear only after a few years [3]. However, at slightly higher radiation doses than applied in our studies in chapters 2, 3 and 4, some have reported a decrease in executive functioning already after six months post radiotherapy in patients with primary brain tumors [4]. Nevertheless, time since radiotherapy was not associated with cognitive functioning in the studies presented in chapters 2, 3 and 4. These studies included patients with long follow-up time since radiotherapy and therefore limited additional damage is expected. Whether the potential radiotherapy induced damage can change over time can only be answered with long-term follow-up studies.

However, even though no differences were found between irradiated and unirradiated patients, NFA patients displayed a worse cognitive performance compared to reference samples. These cognitive impairments can be the consequence of surgery, however, previous studies indicated that it is more likely that patients already exhibit such impairments before surgery. For example, it was demonstrated in patients with frontal meningiomas that surgery did not impair patients' cognitive functioning, since pre- and postoperative assessments revealed no significant differences with regard to cognition [5]. This idea is consistent with a study of Tucha and colleagues who found that more than 90% of patients with brain tumors have cognitive impairments at baseline [6]. Another explanation for impaired cognitive performance of NFA patients might be that nonspecific psychological factors associated with having a chronic illness have a negative effect on cognition [7]. Alternatively, hormonal insufficiency secondary to the tumor and/or its treatment could also be responsible for impaired cognitive functioning of patients with NFA. In particular, growth hormone (GH) and glucocorticoids may play an important role in cognition, as will also be discussed in the following sections.

A strength of the aforementioned studies is the fixed position of the pituitary tumor in NFA patients excluding potential confounding influences of the localization of both brain tumor and subsequent targeted focal radiotherapy. Also, as already mentioned, compared to previous studies a relatively large group of patients with a single diagnosis was included in **chapter 2.**

There are, however, some limitations that should be taken into account. First, the disease characteristics (e.g. larger and more invasive tumors) of patients who receive both surgery and radiotherapy are somewhat different than the disease characteristics of NFA patients who only receive surgery, indicating a possible selection bias. Second, cognition was only assessed with a relatively limited test battery and the studies described in chapters 2, 3 and 4 only focused on memory and executive functioning. A more sensitive approach would be to use several tests to assess the cognitive domains of memory and executive functioning. In addition, other cognitive domains should be examined as well.

The effects of previous growth hormone excess and current medical treatment for acromegaly on cognition

Changes in cognitive functioning can be expected as a consequence of GH excess or GH deficiency [8-12]. The aim of **chapter 5** was two-fold. First, to compare cognitive performance of acromegaly patients with a persistent disease (i.e. on GH suppressive medication) with the performance of acromegaly patients who were in remission. Until now, no research is available on the effects of medical treatment for acromegaly on cognition. Learning more about the association between medical treatment for acromegaly and cognitive performances is important, since studies indicated that GH suppressive medication can lead to changes in diurnal GH profile, despite Insulin-like growth factor-1 (IGF-1) values being within the normal range [13, 14]. The second aim was to compare cognitive performance of acromegaly patients with NFA patients, to investigate the effects of previous GH excess on cognition. So far, most studies on the effects of GH excess on cognition compared untreated acromegaly patients [11] or acromegaly patients who are not fully biochemically controlled [12] with healthy participants. These kinds of comparisons appear to be less informative since psychological factors associated with having a chronic illness might influence the results [7]. Therefore, we compared acromegaly patients with NFA patients, since NFA patients share many disease characteristics with acromegaly patients but do not have GH excess.

Our study showed that acromegaly and NFA patients displayed a decreased cognitive performance compared to reference populations, however, previous GH excess had no effect on cognition. In addition, *Z-scores* did not differ in cognition from both acromegaly patients on GH suppressive medication with normal IGF-1 *Z-scores* compared to both acromegaly patients who were in remission and NFA patients. Although not measured directly, diurnal GH profiles are likely to be different between patient groups in this study. Our findings suggest that disease control by IGF-1 alone might be sufficient to prevent cognitive decline. This conclusion needs, however, to be confirmed in future studies, since our study had a cross-sectional design and was therefore limited to infer causality. Additional studies should also determine GH profiles to see what the actual effects are of possible abnormal GH profiles on cognition.

A limitation of this study was that there were several significant differences between acromegaly and NFA patients regarding their baseline characteristics, which potentially could have influenced cognition. These groups particularly differed with regard to the frequency of radiotherapy, i.e. acromegaly patients with persistent disease received radiotherapy more often than both acromegaly patients who were in remission and NFA patients. Based on the results of the previous chapters and a regression analysis focusing on the effects of radiotherapy on cognition in acromegaly patients, we concluded that the differences in frequency of radiotherapy did not influence the results reported in **chapter 5**. Furthermore, NFA patients displayed significantly lower IGF-1 *Z-scores* compared to the acromegaly patients. Even though NFA patients were probably not GH deficient, it is possible that IGF-1 *Z-scores* closer to zero are more favorable for cognition compared to IGF-1 *Z-scores* that are more than one standard deviation below zero.

We concluded that patients with acromegaly show impairments in memory and executive functioning. However, based on this study, endocrinologists can inform acromegaly patients with an active disease that GH suppressive medication has no additional detectable negative effect on memory and executive functioning. Also, cognition is not negatively influenced by previous GH

excess. This conclusion needs, however, to be confirmed in prospective studies, since our study had a cross-sectional design and was therefore limited to infer causality.

The effects of hydrocortisone substitution on cognition in patients with secondary adrenal insufficiency due to various pituitary diseases

Patients with secondary adrenal insufficiency (SAI), as a consequence of pituitary disease, are treated with glucocorticoids dose regimens that inevitably result in over- or under-replacement during certain periods of the day which can affect cognitive functioning. Deficits in memory, executive functioning, attention and social cognition were expected since these functions rely on brain structures containing high concentrations of glucocorticoids receptors [15, 16]. In **chapter 6** we described the results of the first study on the effects of long-term physiological doses of hydrocortisone and cognitive functioning in patients treated for SAI. It is important to determine these long-term effects, since cognitive impairments usually affect everyday functioning. Our study described in **chapter 6** showed that patients with pituitary diseases all of whom were treated for SAI are selectively impaired in memory, attention, executive functions and social cognition compared to reference samples. These selective impairments might at least in part be determined by the material that has to be processed (verbal versus non-verbal, coherent versus incoherent). Furthermore, various forms of treatment appeared to be associated with cognition in SAI patients. Both, characteristics of radiotherapy and surgery were associated with a poorer memory, executive functioning, attention and social cognition. In **chapter 2** and **chapter 3**, we did not reveal such effects of radiotherapy and surgery when comparing NFA patients to reference samples. Also regarding self-reported cognition these effects were not found [17]. An explanation for this apparent discrepancy might be that a more comprehensive test battery was used in the study described in **chapter 6**, which might be more sensitive in detecting cognitive impairments in pituitary patients.

When a more clinical individual approach was used, it was found that many SAI patients showed impairments in the cognitive domains of memory (80%), attention (85%), executive functioning (43%) and social cognition (37%). These results indicate that SAI patients might benefit from cognitive rehabilitation programs, which are already applied on patients with primary brain tumors [18]. Compensatory techniques can among other benefits be helpful in teaching patients how to manage their daily life, since problems in attention and executive function are also reported in outpatient clinics. Furthermore, treatment can also be directed on improving social cognitive skills to maintain good interpersonal relations. With regard to memory, the results specifically indicated that the consolidation of verbal information appears to be intact in SAI patients, however, these patients do show difficulties with encoding and retrieving information from memory. Furthermore, patients treated for SAI appear to pay more attention to visual stimuli as compared to auditory stimuli. Taking these factors into account, rehabilitation programs in these patients should concentrate on the use of visual cues that can facilitate the retrieval of information from memory.

Conclusions and future directions

The present thesis describes the effects of 1) radiotherapy, 2) GH excess in history and current GH suppressive medication and 3) hydrocortisone substitution therapy on cognition in patients with pituitary diseases. Based on the results of this thesis it can be concluded that:

- Current multiple field radiotherapy techniques and fractionated radiation dose regimens do not appear to have a major effect on memory and executive functioning in patients with NFA.
- Previous GH excess in patients is not negatively associated with cognition.
- Current medical treatment for GH excess is not related to cognition.
- Patients with various pituitary diseases who all were treated for SAI have selective impairments in the cognitive domains of memory, attention, executive functions and social cognition.

To further elucidate the effects of treatment on cognition in patients with pituitary diseases several recommendations can be made. First, regarding the effects of surgery and radiotherapy on cognition it is recommended to perform longitudinal studies with comprehensive test batteries and a baseline measurement before any kind of treatment is started. A study like this will be the only way to distinguish between the effects of disease and treatment related factors on cognition. A longitudinal study including a baseline assessment is also important, since there is a lot of inter-individual variability in the ability to recover after brain pathology. This inter-individual variability has been explained by the construct of "cognitive reserve". Cognitive reserve refers to a set of variables, including education, intelligence, and mental stimulation, which putatively allow the brain to adapt to -and hence mask- underlying pathologies by maintaining cognitive functioning despite underlying neural changes [19]. A consequence might be that patients with pituitary diseases and a high cognitive reserve show less cognitive impairments than patients with pituitary diseases with a low cognitive reserve. Cognitive reserve thus appears to be a factor that should be taken into account in future studies.

Second, when comparing patient data with reference samples, as done in studies in this thesis, it is important to keep in mind that there are differences regarding the way reference data have been collected and the way the data in the studies of this thesis have been collected. For example, there might be an effect of the order in which the tests were applied. Also, more general psychological factors such as fatigue or motivation might be of influence, particularly if long and extensive test batteries are used (e.g. Chapter 6). Future studies on cognition in patients with pituitary diseases should therefore include a control group, which consists of healthy individuals who are matched for age, sex and education to the group of patients.

Finally, it needs to be taken into account that a cross sectional study was used to determine the effects of hydrocortisone on cognition. Cross sectional studies are effective when evaluating large groups of patients and determining the associations with treatment characteristics. However, no conclusions can be drawn about the causal relationship between hydrocortisone exposure and cognitive outcome, since exposure and outcome are determined at the same time. For future research it is therefore recommended to assess and compare in a randomized double blind design the effects of a low physiological hydrocortisone substitution dose with the effects of a high physiological hydrocortisone substitution dose on cognition in order to learn more about

the effects of hydrocortisone (or cortisone acetate) on cognition in patients treated for SAI. This kind of study is also informative regarding the effects of other pituitary hormones on cognition.

In summary, the following findings in our studies are relevant for clinicians who care for pituitary patients:

1. Invariably we found cognitive defects in pituitary patients. Doctors should be aware that this is found frequently in patients visiting the outpatient clinics and may have clear repercussions. Although not the topic of this study consequences of this may be the inability to properly carry out your work, difficulties in managing the day and reduced self confidence. There appears to be a compensatory mechanism (i.e. enhanced visual memory) that enables the patients to adapt to a certain extent. This is a finding that may provide reassurance in patients and potentially provides the opportunity to develop a cognitive rehabilitation program.
2. Clinicians should be aware of safety profiles of treatment modalities. Radiotherapy for pituitary diseases is not negatively associated with major effects on cognition, and this argument should not be used to postpone or withhold radiotherapy to patients who can benefit from it. Further, in our cross sectional studies we found no effects of medical treatment for acromegaly.

References

1 Postma TJ, Klein M, Verstappen CC, Bromberg JE, Swennen M, Langendijk JA, Taphoorn MJ, Scheltens P, Slotman BJ, van der Ploeg HM, Aaronson NK, Heimans JJ 2002 Radiotherapy-induced cerebral abnormalities in patients with low-grade glioma. *Neurology* 59(1): 121-123.

2 Douw L, Klein M, Fagel SS, van den Heuvel J, Taphoorn MJ, Aaronson NK, Postma TJ, Vandertop WP, Mooij JJ, Boerman RH, Beute GN, Sluimer JD, Slotman BJ, Reijneveld JC, Heimans JJ 2009 Cognitive and radiological effects of radiotherapy in patients with low-grade glioma: Long-term follow-up. *Lancet Neurol* 8(9): 810-818.

3 Armstrong CL, Hunter JV, Ledakis GE, Cohen B, Tallent EM, Goldstein BH, Tochner Z, Lustig R, Judy KD, Pruitt A, Mollman JE, Stanczak EM, Jo MY, Than TL & Phillips P 2002 Late cognitive and radiographic changes related to radiotherapy: Initial prospective findings. *Neurology* 59(1): 40-48.

4 Hahn CA, Zhou SM, Raynor R, Tisch A, Light K, Shafman T, Wong T, Kirkpatrick J, Turkington T, Hollis D & Marks LB. 2009 Dose-dependent effects of radiation therapy on cerebral blood flow, metabolism, and neurocognitive dysfunction. *Inter J Rad Oncol, Biol, Phys.* 73(4): 1082-1087.

5 Tucha O, Smely C, Preier M, Becker G, Paul GM, Lange KW 2003 Preoperative and postoperative functioning in patients with frontal meningiomas *J Neurosurg* 98(1): 21-31.

6 Tucha O, Smely C, Preier M and Lange KW 2000 Cognitive deficits before treatment among patients with brain tumors. *Neurosurgery* 47(2): 324-334.

7 van den Berg E., Kloppenburg, RP, Kessels RP, Kappelle LJ, Biesels GJ 2009 Type 2 diabetes mellitus, hypertension, dyslipidemia and obesity: a systematic comparison of their impact on cognition. *Biochim Biophysi Acta*, 1792(5): 470-481.

8 Sathiavageeswaran M, Burman P, Lawrence D, Harris AG, Falleti MG, Maruff P 2007 Effects of GH on cognitive function in elderly patients with adult-onset GH deficiency: a placebo-controlled 12-month study. *Eur J Endocrinol* 156(4): 439-447.

9 Oertel H, Schneider HJ, Stalla K, Holsboer F, Zihl J 2004 The effect of growth hormone substitution on cognitive performance in adult patients with hypopituitarism. *Psychoneuroendocrinology* 29(7): 839-850.

10 Faletti MG, Maruff P, Burman P, Harris A 2006 The effects of growth hormone (GH) deficiency and GH replacement on cognitive performance in adults: a meta analysis of the current literature. *Psychoneuroendocrinology* 31(6): 681-691.

11 Leon-Carrion J, Martin-Rodriguez JF, Madrazo-Atutxa A, Soto-Moreno A, Venegas-Moreno A, Torres-Vela E, Benito-López P, Gálvez MA, Tinahones FJ, Leal-Cerro A. 2010 Evidence of cognitive and neurophysiological impairment in patients with untreated naive acromegaly. *J Clin Endocrinolo Metab* 95(9): 4367-4379.

12 Sievers C, Sämann PG, Pfister H, Dimopoulou C, Czisch M, Roemmler J, Schopohl J, Stalla GK, Zihl J. 2012 Cognitive function in acromegaly: description and brain volumetric correlates *Pituitary* 15(3): 350-357.

13 Biermasz NR, Pereira AM, Frölich M, Romijn JA, Veldhuis JD, Roelfsema F 2004 Ocreotide represses secretory-burst mass and nonpulsatile secretion but does not restore event frequency or orderly GH secretion in acromegaly *Am J Physiol Endocrinol Metab* 286: E25-E30.

14 Neggers SJ, Kopchick JJ, Jørgensen, van der Lely AJ 2011 Hypothesis: Extra-hepatic acromegaly: a new paradigm? *Europ J Endocrinol* 164(1): 11-16.

15 Joels M 2008 Functional actions of corticosteroids in the hippocampus. *Eur J Pharmacol* 583(2- 3): 312-321.

16 Sanchez MM, Young LJ, Plotsky PM, Insel TR 2000 Distribution of corticosteroid receptors in the rhesus brain: Relative absence of glucocorticoid receptors in the hippocampal formation. *J Neurosci* 20(12): 4657-4668.

17 van Beek AP, van den Bergh AC, van den Berg LM, van den Berg G, Keers JC, Langendijk JA, Wolffenbuttel BH 2007 Radiotherapy is not associated with reduced quality of life and cognitive function in patients treated with nonfunctioning pituitary adenoma. *Int J Radiat Oncol Biol Phys* 68(4): 986-991.

18 Zuchella C, Capone A, Codella V, De Nunzio AM, Vecchione C, Sandrini G, Pace A, Pierelli F, Bartolo M 2013 Cognitive rehabilitation for early post-surgery inpatients affected by primary brain tumor: a randomized, controlled trial. *J Neuro oncol* 114(1): 93-100.

19 Robertson IH 2013 A noradrenergic theory of cognitive reserve: implications for Alzheimer's disease. *Neurobiol of Aging* 34(1): 298–308.

Chapter 8

Nederlandse samenvatting

Samenvatting

In dit proefschrift staan de resultaten van studies naar de effecten van verschillende behandelmogelijkheden op het cognitieve functioneren van patiënten met hypofyse aandoeningen beschreven. Betrouwbare informatie over de mogelijke effecten van verschillende behandelingen is belangrijk voor clinici en patiënten zodat zij een geïnformeerde discussie kunnen voeren en beslissing kunnen nemen. De kennis over de impact van specifieke behandelingen op het cognitieve functioneren zou tevens kunnen dienen als een basis voor een meer theoretische evaluatie van verschillende behandelopties.

De effecten van radiotherapie op cognitie bij patiënten die zijn behandeld voor een niet functionerend hypofyse adenoom

Het doel van **hoofdstuk 2** was om het geheugen en het executief functioneren te beoordelen in een grote groep patiënten (n = 84) die behandeld zijn voor een niet functionerend hypofyse adenoom (NFA). We hebben ons gefocust op deze aandoening omdat de wisselende uitkomsten van verschillende studies zeer waarschijnlijk zijn veroorzaakt door de inclusie van kleine en heterogene patiënten populaties. Bovendien hebben patiënten met een NFA het voordeel dat zij geen voorgeschiedenis kennen met daarin een overmatige productie van één bepaald hormoon. Dit laatste is bijvoorbeeld wel het geval bij patiënten met de ziekte van Cushing, waarbij een teveel aan cortisol (stresshormoon) het cognitief functioneren kan beïnvloeden. Het tweede doel in dit hoofdstuk was om patiënten die alleen een chirurgische behandeling hebben ontvangen te vergelijken met patiënten die aanvullend aan de chirurgie ook radiotherapie hebben gekregen. De resultaten van het onderzoek lieten zien dat de groep NFA patiënten in zijn geheel slechter scoorde op zowel de verbale geheugentest als ook op de executieve functie test wanneer hun testprestaties werden vergeleken met testprestaties van mensen uit referentie populaties. De effecten van radiotherapie op het cognitieve functioneren lijken hierbij zeer klein te zijn en zijn waarschijnlijk klinisch niet relevant.

Echter, omdat kleine effecten van radiotherapie op cognitie bij NFA patiënten niet kunnen worden uitgesloten (bijv. vanwege het gebruik van verschillende radiotherapeutische technieken en daardoor verschillende dosesverdelingen in het brein), werd besloten om de onderzoeksmethode te verfijnen. In **hoofdstuk 3** hebben we gekeken of er een relatie bestaat tussen de hoeveelheid bestraling op een specifiek hersengebied en de cognitieve functie die een beroep doet op dat specifieke hersengebied. Als hersengebieden werden de hippocampus en de prefrontale cortex gekozen, gebieden die belangrijk zijn voor respectievelijk het geheugen en de executieve functies. Dit vervolgonderzoek liet zien dat de huidige radiotherapeutische technieken geen negatieve invloed lijken te hebben op het geheugen en het executief functioneren bij patiënten die zijn behandeld voor een NFA. Tevens vonden wij geen relatie tussen de hoeveelheid bestraling op de hippocampus en het geheugen en ook niet tussen de hoeveelheid bestraling op de prefrontale cortex en het executief functioneren. De radiotherapeutische technieken zoals deze staan beschreven in hoofdstuk 2 en 3 lijken derhalve veilig te zijn.

Radiotherapie wordt ook in verband gebracht met hersenafwijkingen zoals witte stof schade

en cerebrale atrofie welke geassocieerd worden met een verminderd cognitief functioneren. Tot dusver hebben de beeldvormende studies naar de associaties tussen deze structurele hersenafwijkingen en het cognitief functioneren gebruik gemaakt van patiënten met verschillende tumor lokalisaties. Dit waren meestal patiënten met laaggradige gliomen waarbij verschillende cognitieve domeinen zijn aangedaan vanwege verschillende lokalisaties van de tumor in het brein wat leidt tot wisselende resultaten. In het in **hoofdstuk 4** beschreven onderzoek werd er daarom voor gekozen om structurele hersenafwijkingen te beoordelen in relatie tot het cognitieve functioneren bij patiënten die zijn behandeld voor een NFA aangezien bij deze patiënten dus sprake is van een gefixeerde tumor lokalisatie. De resultaten van deze studie lieten zien dat de frequentie van structurele hersenafwijkingen niet verschilt tussen patiënten met beperkingen in de cognitieve domeinen geheugen en executief functioneren en patiënten zonder beperkingen in het geheugen en het executief functioneren.

Kortom, de resultaten van de hoofdstukken 2, 3 en 4 suggereren dat de huidige radiotherapie technieken en de gefractioneerde bestralingsplannen die NFA patiënten krijgen in aanvulling op hun chirurgische behandeling geen groot negatief effect lijken te hebben op het geheugen en het executief functioneren.

Eén van de mogelijke verklaringen voor onze bevindingen is dat er sprake is van een soort dosis gerelateerde drempelwaarde voor radiotherapie voordat deze leidt tot cognitieve beperkingen, wat suggereert dat alleen boven die drempelwaarde een dosis-respons relatie kan worden gevonden. De resultaten van de hoofdstukken 2, 3 en 4 suggereren dat alle radiotherapeutische technieken en gefractioneerde bestralingsplannen die in de afgelopen decennia zijn gebruikt voor de behandeling van NFA patiënten in ons centrum onder deze drempelwaarde lijken te opereren en daardoor geen grote negatieve gevolgen lijken te hebben voor het geheugen en het executief functioneren.

Een andere mogelijkheid is dat de nadelige effecten van radiotherapie nog niet tot uiting zijn gekomen en dat cognitieve beperkingen pas tot uiting komen na een aantal jaren. Hier pleit echter tegen dat andere onderzoekers bij enigszins hogere radiotherapeutische doseringen dan die in onze studies werden onderzocht, al zes maanden na de radiotherapie cognitief verval rapporteerden bij patiënten met primaire hersentumoren. In de studies die staan beschreven in de hoofdstukken 2, 3 en 4 was de tijd sinds radiotherapie niet geassocieerd met het cognitief functioneren. Aangezien in onze studies patiënten zijn geïncludeerd met een lange follow-up tijd sinds radiotherapie, verwachten wij geringe aanvullende schade. Of deze potentiële radiotherapeutisch geïnduceerde schade kan veranderen over tijd kan alleen worden beantwoord met behulp van lange termijn follow-up studies.

Hoewel er geen verschillen werden gevonden tussen de bestraalde en niet-bestraalde patiënten, lieten de NFA patiënten wel verminderde cognitieve prestaties zien in vergelijking met de testprestaties van mensen uit referentie populaties. Deze cognitieve beperkingen kunnen het gevolg zijn van de chirurgische ingreep, maar eerdere studies suggereren dat het waarschijnlijker is dat patiënten reeds cognitieve beperkingen vertoonden voorafgaand aan de operatie. Zo werd bijvoorbeeld bij mensen met frontale meningeomen aangetoond dat een operatie waarschijnlijk

niet van invloed was op het cognitief functioneren, omdat pre- en postoperatieve cognitieve prestaties niet significant van elkaar verschilden. Deze suggestie is in overeenstemming met een studie van Tucha en collega's die vonden dat meer dan 90% van de patiënten met hersentumoren cognitieve beperkingen laat zien tijdens een baseline bepaling. Een andere verklaring voor de verminderde cognitieve prestaties van NFA patiënten zou kunnen zijn dat aspecifieke psychologische factoren, die geassocieerd zijn met het hebben van een chronische ziekte, een negatief effect hebben op het cognitieve functioneren. Ten slot kan het ook zo zijn dat de hormonale insufficiëntie secundair aan de tumor en/of de behandeling verantwoordelijk is voor een verminderd cognitief functioneren van patiënten met een NFA. In het bijzonder kunnen het groeihormoon (GH) en glucocorticoïden van invloed zijn op het cognitief functioneren, zoals wordt besproken in de volgende paragrafen.

Eén van de sterke punten van de bovengenoemde studies is de eenduidige lokalisatie van de hypofyse tumor bij NFA patiënten met als gevolg dat mogelijke verstorende invloeden van de lokalisatie van zowel hersentumor als ook de daaropvolgende gerichte focale radiotherapie kunnen worden uitgesloten. Bovendien, zoals al eerder werd vermeld, is een relatief grote homogene groep patiënten geïncludeerd in hoofdstuk 2.

Er zijn echter ook zwakke punten waarmee rekening moet worden gehouden. Ten eerste, de ziekte kenmerken (bv. grotere en meer invasieve tumoren) van patiënten die zowel chirurgie als radiotherapie ontvangen, zijn anders dan de ziekte kenmerken van NFA patiënten die alleen chirurgie ontvangen, wat wijst op een mogelijke selectiebias. Ten tweede, het cognitieve functioneren werd alleen met een relatief beperkte testbatterij onderzocht en de studies zoals beschreven in de hoofdstukken 2, 3 en 4 beoordeelden alleen het geheugen en het executief functioneren. Een gevoeligere methode zou zijn om verschillende tests te gebruiken om de cognitieve domeinen geheugen en executieve functioneren te beoordelen. Daarnaast is het van belang dat er ook onderzoek wordt gedaan naar andere cognitieve domeinen.

De effecten van een teveel aan groeihormoon in de voorgeschiedenis en de huidige medische behandeling voor acromegalie op cognitie

Veranderingen in het cognitief functioneren kunnen het gevolg zijn van een overmaat aan GH of juist een GH deficiëntie. Het doel van **hoofdstuk 5** was tweeledig. Ten eerste werden de cognitieve prestaties van acromegalie patiënten met een aanhoudende ziekte (d.w.z. op GH onderdrukkende medicatie) vergeleken met de cognitieve prestaties van acromegalie patiënten die in remissie waren. Tot op heden is er geen onderzoek gedaan naar de effecten van de medische behandeling van acromegalie op het cognitieve functioneren. Het is echter belangrijk om meer te leren over de associatie tussen de medische behandeling voor acromegalie en cognitieve prestaties, omdat eerdere studies aantonen dat GH onderdrukkende medicatie kan leiden tot veranderingen in het dagelijkse GH profiel, ondanks dat de waarde van het Insuline gelijkende groeifactor (IGF-1) binnen de normale range ligt. Het tweede doel van **hoofdstuk 5** was om de cognitieve prestaties van acromegalie patiënten te vergelijken met de cognitieve prestaties van NFA patiënten zodat de gevolgen van een overmaat aan GH in de voorgeschiedenis konden worden onderzocht. Tot dusver vergeleken de meeste studies naar de effecten van een overmaat aan GH op het

cognitief functioneren onbehandelde acromegalie patiënten of acromegalie patiënten die niet volledig biochemisch gecontroleerd waren met gezonde vrijwilligers. Dergelijke vergelijkingen lijken minder informatief te zijn, omdat psychologische factoren die geassocieerd zijn met het hebben van een chronische ziekte de resultaten kunnen beïnvloeden. Om deze reden hebben wij acromegalie patiënten met NFA patiënten vergeleken, omdat NFA patiënten veel kenmerken met acromegalie patiënten delen, maar geen overmaat aan GH in de voorgeschiedenis kennen.

De resultaten van **hoofdstuk 5** toonden aan dat acromegalie en NFA patiënten beide verminderde cognitieve prestaties lieten zien in vergelijking met referentiepopulaties en dat een voorgeschiedenis van een overmaat aan GH geen blijvend effect lijkt te hebben op het cognitieve functioneren. Bovendien verschilden de prestaties op de cognitieve tests van acromegalie patiënten op GH onderdrukkende medicatie (met IGF-1 scores die in de normale range liggen) niet van de prestaties van zowel acromegalie patiënten die in remissie waren als van NFA patiënten. Hoewel niet direct gemeten, verschilden de dagelijkse GH profielen waarschijnlijk tussen de groepen patiënten die onderzocht zijn in onze studie in **hoofdstuk 5**. Onze bevindingen suggereren dat de controle van de ziekte door alleen IGF-1 voldoende zou kunnen zijn om cognitieve beperkingen te voorkomen. Deze conclusie moet echter worden bevestigd in toekomstige studies vanwege het cross-sectionele design van onze studie waardoor we geen uitspraak kunnen doen over de causaliteit. Aanvullende onderzoeken zouden ook GH profielen moeten bepalen om te zien wat de werkelijke gevolgen zijn van eventuele afwijkende GH profielen op het cognitieve functioneren.

Een beperking van dit onderzoek was dat er een aantal belangrijke baseline verschillen waren tussen de acromegalie patiënten en de NFA patiënten, die mogelijk een invloed hebben gehad op het cognitieve functioneren. Deze groepen verschilden vooral wat betreft het krijgen van radiotherapie; de acromegalie patiënten met een persisterende ziekte kregen vaker radiotherapie dan de acromegalie patiënten die in remissie waren en de NFA patiënten. Op basis van de resultaten van de voorgaande hoofdstukken en een regressieanalyse naar de effecten van radiotherapie op cognitie bij acromegalie patiënten, kan worden aangenomen dat het verschil in frequentie van radiotherapie de resultaten zoals beschreven in **hoofdstuk 5** niet heeft beïnvloed. Bovendien hadden de NFA patiënten significant lagere IGF-1 Z-scores in vergelijking met de acromegalie patiënten. Hoewel de NFA patiënten waarschijnlijk niet GH deficiënt waren, suggereert onderzoek van anderen dat IGF-1 scores die dichter bij het gemiddelde van de normale range liggen gunstiger zijn voor het cognitieve functioneren dan IGF-1 scores die juist meer afwijken van het gemiddelde in de normale range.

We kunnen dus concluderen dat patiënten met acromegalie een verminderde prestatie laten zien op het gebied van het geheugen en het executieve functioneren. Echter, op basis van deze studie, kunnen endocrinologen acromegalie patiënten met een actieve ziekte informeren over het feit dat GH onderdrukkende medicatie geen additionele aantoonbare beperkingen in het geheugen en het executief functioneren tot gevolg heeft. Ook lijkt het cognitieve functioneren niet negatief te worden beïnvloed door een overmaat aan GH in de voorgeschiedenis. Deze conclusie moet echter worden bevestigd in prospectieve studies.

De effecten van hydrocortison substitutie op cognitie bij patiënten met secundaire bijnierschorsinsufficiëntie door verschillende hypofyse aandoeningen

Patiënten met secundaire bijnierschorsinsufficiëntie, als gevolg van een hypofyse-aandoening, worden behandeld met verschillende doseringen glucocorticoïden die onvermijdelijk leiden tot over- of ondersubstitutie gedurende bepaalde perioden van de dag. Deze over- of ondersubstitutie kan van invloed zijn op het cognitief functioneren. Veranderingen in het geheugen, de executieve functies, de aandacht en de sociale cognitie kunnen worden verwacht, aangezien deze cognitieve functies een beroep doen op hersenstructuren die hoge concentraties glucocorticoïden-receptoren bevatten. In **hoofdstuk 6** worden de resultaten beschreven van de eerste studie naar het cognitief functioneren van patiënten die langdurig zijn behandeld voor secundaire bijnierschorsinsufficiëntie met fysiologische doseringen hydrocortison. De resultaten van de studie laten zien dat patiënten met hypofyse aandoeningen – die allen werden behandeld voor secundaire bijnierschorsinsufficiëntie - selectieve beperkingen laten zien in het geheugen, de aandacht, de executieve functies en de sociale cognitie in vergelijking met referentie populaties van gezonde personen. Deze selectieve stoornissen lijken gedeeltelijk te worden bepaald door het materiaal dat moet worden verwerkt (verbaal versus non-verbaal, coherent versus incoherent). Bovendien lijken verschillende behandelmogelijkheden geassocieerd te zijn met het cognitieve functioneren van patiënten met secundaire bijnierschorsinsufficiëntie. Zowel kenmerken van radiotherapie als ook van chirurgie zijn geassocieerd met een verminderd geheugen, executief functioneren, aandacht en sociale cognitie. In hoofdstuk 2 en hoofdstuk 3 hebben we dergelijke effecten van radiotherapie en chirurgie niet gevonden bij het vergelijken van NFA patiënten met de testprestaties van mensen uit referentie populaties. Ook ten aanzien van zelfgerapporteerde cognitieve beperkingen werden deze effecten niet gevonden. Een verklaring voor deze schijnbare discrepantie zou kunnen zijn dat een meer uitgebreide testbatterij werd gebruikt in de studie zoals beschreven in **hoofdstuk 6**, die mogelijk gevoeliger is in het opsporen van cognitieve beperkingen bij patiënten met hypofyse aandoeningen.

Wanneer meer op individueel niveau werd gekeken, dan zien we dat veel patiënten met secundaire bijnierschorsinsufficiëntie beperkingen vertonen in het geheugen (80%), de aandacht (85%), het executief functioneren (43%) en de sociale cognitie (37%). Deze resultaten laten zien dat deze patiënten mogelijk kunnen profiteren van cognitieve revalidatieprogramma's. Dergelijke programma's worden al toegepast bij patiënten met primaire hersentumoren. Het aanleren van compensatie technieken kan onder andere nuttig zijn om patiënten te helpen om hun dagelijks leven goed te beheren, vooral omdat problemen in de aandacht en de executieve functies (bijv. planning) gemeld worden tijdens poliklinische bezoeken. Bovendien kan de behandeling ook worden gericht op het verbeteren van sociaal cognitieve vaardigheden zodat goede interpersoonlijke relaties kunnen worden onderhouden. De resultaten van de geheugentests laten zien dat de consolidatie van verbale informatie intact lijkt te zijn bij patiënten met secundaire bijnierschorsinsufficiëntie, maar dat deze patiënten problemen hebben met het inprenten en ophalen van informatie uit het geheugen. Bovendien lijken de patiënten meer aandacht te besteden aan visuele stimuli in vergelijking met auditieve stimuli. Revalidatie programma's zouden hier reke-

ning mee kunnen houden door meer gebruik te maken van visuele aanwijzingen die het ophalen van informatie uit het geheugen kunnen vergemakkelijken.

Conclusies en aanbevelingen voor de toekomst

In dit proefschrift worden de resultaten beschreven van 1) radiotherapie, 2) een overmaat aan GH in de voorgeschiedenis en van GH onderdrukkende medicatie en 3) hydrocortison substitutie therapie (als onderdeel van vaak meervoudige hormonale substitutietherapie) in relatie tot cognitie bij patiënten die behandeld zijn voor hypofyse aandoeningen. Op basis van dit proefschrift kunnen we het volgende concluderen:

- De huidige radiotherapie technieken en gefractioneerde bestralingsplannen lijken geen grote invloed te hebben op het geheugen en het executief functioneren bij patiënten met een NFA.
- Een voorgeschiedenis van een overmaat aan GH lijkt niet schadelijk te zijn voor het cognitieve functioneren.
- De huidige medische behandeling voor een overmaat aan GH lijkt niet gerelateerd te zijn aan het cognitieve functioneren.
- Patiënten met verschillende hypofyse aandoeningen, die allen werden behandeld voor secundaire bijnierschorsinsufficiëntie, hebben selectieve stoornissen in de cognitieve domeinen van het geheugen, de aandacht, de executieve functies en de sociale cognitie.

Meerdere aanbevelingen kunnen worden gedaan om de effecten van behandeling op het cognitieve functioneren bij patiënten met hypofyse aandoeningen verder te ontrafelen. Ten eerste adviseren we om - met betrekking tot de effecten van chirurgie en radiotherapie op het cognitieve functioneren - longitudinale studies uit te voeren met uitgebreide test batterijen en een baseline meting voordat gestart wordt met behandelen. Met een dergelijk onderzoek kan onderscheid gemaakt worden tussen de effecten van de ziekte enerzijds en de effecten van behandeling op cognitie anderzijds. Een longitudinaal onderzoek inclusief een baseline bepaling is tevens belangrijk, omdat er veel interindividuele variabiliteit is in het vermogen om te herstellen nadat er schade is ontstaan aan het brein. Deze interindividuele variabiliteit is mogelijk te verklaren door het construct "cognitieve reserve". Dit verwijst naar een set van variabelen, waaronder onderwijs, intelligentie en mentale stimulatie, die vermoedelijk mensen in staat stellen om zich aan te passen aan de nieuwe situatie - en daarmee te kunnen compenseren voor een onderliggende pathologie. Het cognitief functioneren kan dan behouden worden, ondanks achterliggende neurale veranderingen. Een gevolg hiervan zou kunnen zijn dat patiënten met hypofyse aandoeningen en een hoge cognitieve reserve minder cognitieve stoornissen tonen dan patiënten met hypofyse aandoeningen en een lage cognitieve reserve. Het construct van cognitieve reserve lijkt dus een factor waarmee rekening moet worden gehouden in toekomstige studies.

Ten tweede, bij het vergelijken van patiëntgegevens met testprestaties van referentie populaties, zoals gedaan in studies in dit proefschrift, is het belangrijk om in gedachten te houden dat er verschillen zijn ten aanzien van de manier waarop referentiegegevens zijn verzameld en de manier waarop de gegevens in de studies van dit proefschrift zijn verzameld. Zo kan er een effect

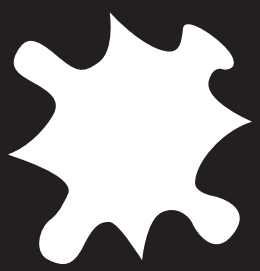
zijn van de volgorde waarin de tests zijn afgenomen. Ook kunnen meer algemene psychologische factoren zoals vermoeidheid of motivatie van invloed zijn, vooral als lange en uitgebreide testbatterijen worden gebruikt (bijv. hoofdstuk 6). Toekomstige studies gericht op het cognitieve functioneren van patiënten met hypofyse aandoeningen zouden daarom ook een controlegroep moeten includeren, die bestaat uit gezonde individuen die zijn gematcht voor leeftijd, geslacht en opleiding aan de patiëntengroep.

Ten slotte moet er rekening worden gehouden met het feit dat er een cross-sectioneel onderzoek werd gedaan naar de effecten van hydrocortison op cognitie. Cross-sectionele studies zijn effectief bij het evalueren van grote groepen patiënten en het bepalen van de associaties met behandelkarakteristieken. Er kunnen echter geen conclusies worden getrokken over het causale verband tussen hydrocortison gebruik en cognitief functioneren, omdat deze op hetzelfde moment zijn bepaald. Voor toekomstig onderzoek wordt daarom geadviseerd om een gerandomiseerd dubbelblind onderzoek te doen naar de effecten van een lage en een hoge fysiologische hydrocortison dosering op het cognitief functioneren. Op deze manier kunnen we meer leren over de effecten van verschillende doseringen hydrocortison (of cortisonacetaat) op het cognitieve functioneren van patiënten die zijn behandeld voor secundaire bijnierschorsinsufficiëntie. Dergelijk onderzoek zou ook informatief kunnen zijn om inzicht te krijgen in de relatie tussen andere hypofyse hormonen en cognitie.

Samenvattend kunnen de volgende bevindingen in onze studies relevant zijn voor clinici die zorg dragen voor hypofyse patiënten:

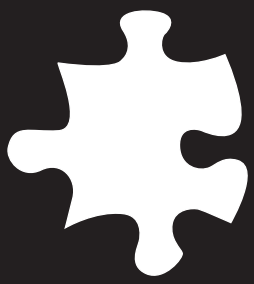
1. Cognitieve tekorten worden vaak gevonden bij de patiënten met hypofyse aandoeningen. Artsen zouden zich hiervan bewust moeten zijn, omdat dit duidelijke gevolgen kan hebben voor de patiënt. Gevolgen van dit verminderd cognitief functioneren kunnen betrekking hebben op het niet goed meer kunnen uitvoeren van het werk, moeilijkheden bij het plannen van de dag of het hebben van een verminderd zelfvertrouwen. Er lijken echter compensatiemogelijkheden aanwezig te zijn waarmee de patiënt zich tot op zeker hoogte kan aanpassen. Dit is een bevinding die geruststelling kan bieden aan patiënten en het zou een nieuw uitgangspunt kunnen zijn bij de ontwikkeling van cognitieve revalidatieprogramma's.
2. Artsen moeten zich bewust zijn van de veiligheidsprofielen van verschillende behandelvormen. Radiotherapie voor hypofyse aandoeningen wordt niet geassocieerd met grote negatieve effecten op het cognitief functioneren en dit argument mag daarom niet worden gebruikt om radiotherapie uit te stellen of te weigeren bij patiënten die hier profijt van kunnen hebben. Verder vonden we in onze cross-sectionele studies geen effecten van een medische behandeling voor acromegalie op het cognitieve functioneren.

About the author



Pauline Brummelman werd op 20 juli 1986 geboren te Eefde. In 2004 behaalde zij haar VWO diploma aan het Isendoorn College te Warnsveld. In hetzelfde jaar startte zij met de studie Psychologie aan de faculteit Gedrags- en Maatschappijwetenschappen van de Rijksuniversiteit Groningen. In 2009 behaalde zij haar mastertitel in de Psychologie met als afstudeerrichting Klinische Neuropsychologie. Haar scriptie beschreef de effecten van radiotherapie op cognitie bij mensen die behandeld zijn voor een hypofyse adenoom. Hierop aansluitend startte zij als promovenda bij de afdeling Endocrinologie van het Universitair Medisch Centrum Groningen onder leiding van dr. A.P. van Beek en Prof. dr. B.H.R. Wolffenbuttel in samenwerking met dr. J. Koerts en Prof. dr. O. Tucha van de afdeling Neuropsychologie van de Rijksuniversiteit Groningen. Tijdens dit promotietraject heeft zij haar bevindingen op vele nationale en internationale congressen gepresenteerd waarbij haar meerdere reisbeurzen zijn toegekend. De resultaten van deze onderzoeken staan beschreven in dit proefschrift. Tijdens haar promotie heeft zij masterthese studenten mogen begeleiden in het afnemen van neuropsychologisch onderzoek en het schrijven van een scriptie. Tevens heeft zij in een relatief korte tijd een interventiestudie opgezet, uitgevoerd en de resultaten daarvan geanalyseerd. In de nabije toekomst zal zij betrokken blijven bij de beschrijving van de hieruit voortgekomen bevindingen. Pauline heeft de ambitie om in haar verdere carrière haar wetenschappelijke kennis te combineren met klinische ervaring om zo een breed inzetbaar psycholoog te worden.

Publications and presentations



Publications

Brummelman P, Sattler MG, Meiners LC, van den Berg G, van der Klauw MM, Elderson MF, Dullaart RP, Koerts J, Tucha O, Wolffenbuttel BH, van den Bergh AC, van Beek AP. Cognitive performance and brain abnormalities on MRI in patients treated for nonfunctioning pituitary macroadenoma. *Submitted* 2013.

Brummelman P, Koerts J, Dullaart RP, van den Berg G, van der Klauw MM, Tucha O, Wolffenbuttel BH, van Beek AP. Cognitive functioning in pituitary patients treated for adrenal insufficiency. *Submitted* 2013.

Brummelman P, Koerts J, Dullaart RP, van den Berg G, Tucha O, Wolffenbuttel BH, van Beek AP. Effects of previous growth hormone excess and current medical treatment for acromegaly on cognition. *European Journal of Clinical Investigation* 2012; 42(12): 1317-1324.

Brummelman P, Sattler MG, Meiners LC, Elderson MF, Dullaart RP, van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van den Bergh AC, van Beek AP. Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex. *European Journal of Endocrinology* 2012; 166(2): 171-179.

Brummelman P, Elderson MF, Dullaart RP, Van den Bergh AC, Timmer CA, van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van Beek AP. Cognitief functioneren bij patiënten die behandeld zijn voor een niet functionerende hypofysemacroadenoom en de effecten van hypofysebestraling. *Neuropraxis* 2011; 15(2): 49-52.

Brummelman P, Elderson MF, Dullaart RP, Van den Bergh AC, Timmer CA, van den Berg G, Koerts J, Tucha O, Wolffenbuttel BH, van Beek AP. Cognitive functioning in patients treated for a nonfunctioning pituitary adenoma and the effects of pituitary radiotherapy. *Clinical Endocrinology* 2011; 74(4): 481-487.

Presentations

Cognitive functioning in patients with secondary adrenal insufficiency. *Mid-year meeting International Neuropsychological Society.* Amsterdam, The Netherlands 2013.

Neurocognitieve gevolgen van niet functionerende hypofyse tumoren en de behandeling hiervan middels radiotherapie. *NVvO Oncologiedag*, Utrecht, The Netherlands 2013.

Cognitive functioning in patients with secondary adrenal insufficiency. *Dutch Endocrine Meeting, Noordwijkerhout*, The Netherlands 2013.

Effects of previous growth hormone excess and current medical treatment for acromegaly on cognition. *European Congress of Endocrinology,* Florence, Italy 2012.

Psychologische aspecten van hypofyse tumoren: voor en na behandeling.
Regionale hypofyse bijeenkomst, Groningen, The Netherlands 2012.

Effects of previous growth hormone excess and current medical treatment for acromegaly on cognition. *Dutch Endocrine Meeting*, Noordwijkerhout, The Netherlands 2012.

Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex. *Annual Meeting of the Endocrine Society,* Boston, US 2011.

Cognitive performance after postoperative pituitary radiotherapy: a dosimetric study of the hippocampus and the prefrontal cortex. *Meeting of Northern-European Neuro-endocrine Group*, Amsterdam, The Netherlands 2011.

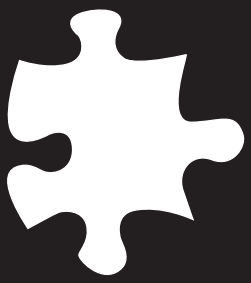
Cognitive functioning in patients treated for a nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Dutch Endocrine Meeting,* Noordwijkerhout, The Netherlands 2011.

Effects of pituitary radiotherapy in patients with a nonfunctioning pituitary macroadenoma. *Annual Meeting of the Endocrine Society*, San Diego, US 2010.

Cognitive functioning in patients treated for a nonfunctioning pituitary macroadenoma and the effects of pituitary radiotherapy. *Young Active Research Club*, Düsseldorf, Germany 2010.

Effects of pituitary radiotherapy in patients with a nonfunctioning pituitary macroadenoma. *Dutch Endocrine Meeting*, Noordwijkerhout, The Netherlands 2010.

Dankwoord



Dankwoord

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Pauline

Pauline Brummelman werd op 20 juli 1986 geboren te Eefde. In 2004 behaalde zij haar VWO diploma aan het Isendoorn College te Warnsveld. In hetzelfde jaar startte zij met de studie Psychologie aan de faculteit Gedrags- en Maatschappijwetenschappen van de Rijksuniversiteit Groningen. In 2009 behaalde zij haar mastertitel in de Psychologie met als afstudeerrichting Klinische Neuropsychologie. Haar scriptie beschreef de effecten van radiotherapie op cognitie bij mensen die behandeld zijn voor een hypofyse adenoom. Hierop aansluitend startte zij als promovenda bij de afdeling Endocrinologie van het Universitair Medisch Centrum Groningen onder leiding van dr. A.P. van Beek en Prof. dr. B.H.R. Wolffenbuttel in samenwerking met dr. J. Koerts en Prof. dr. O. Tucha van de afdeling Neuropsychologie van de Rijksuniversiteit Groningen. Tijdens dit promotietraject heeft zij haar bevindingen op vele nationale en internationale congressen gepresenteerd waarbij haar meerdere reisbeurzen zijn toegekend. De resultaten van deze onderzoeken staan beschreven in dit proefschrift. Tijdens haar promotie heeft zij masterthese studenten mogen begeleiden in het afnemen van neuropsychologisch onderzoek en het schrijven van een scriptie. Tevens heeft zij in een relatief korte tijd een interventiestudie opgezet, uitgevoerd en de resultaten daarvan geanalyseerd. In de nabije toekomst zal zij betrokken blijven bij de beschrijving van de hieruit voortgekomen bevindingen. Pauline heeft de ambitie om in haar verdere carrière haar wetenschappelijke kennis te combineren met klinische ervaring om zo een breed inzetbaar psycholoog te worden.